Fig. 1.

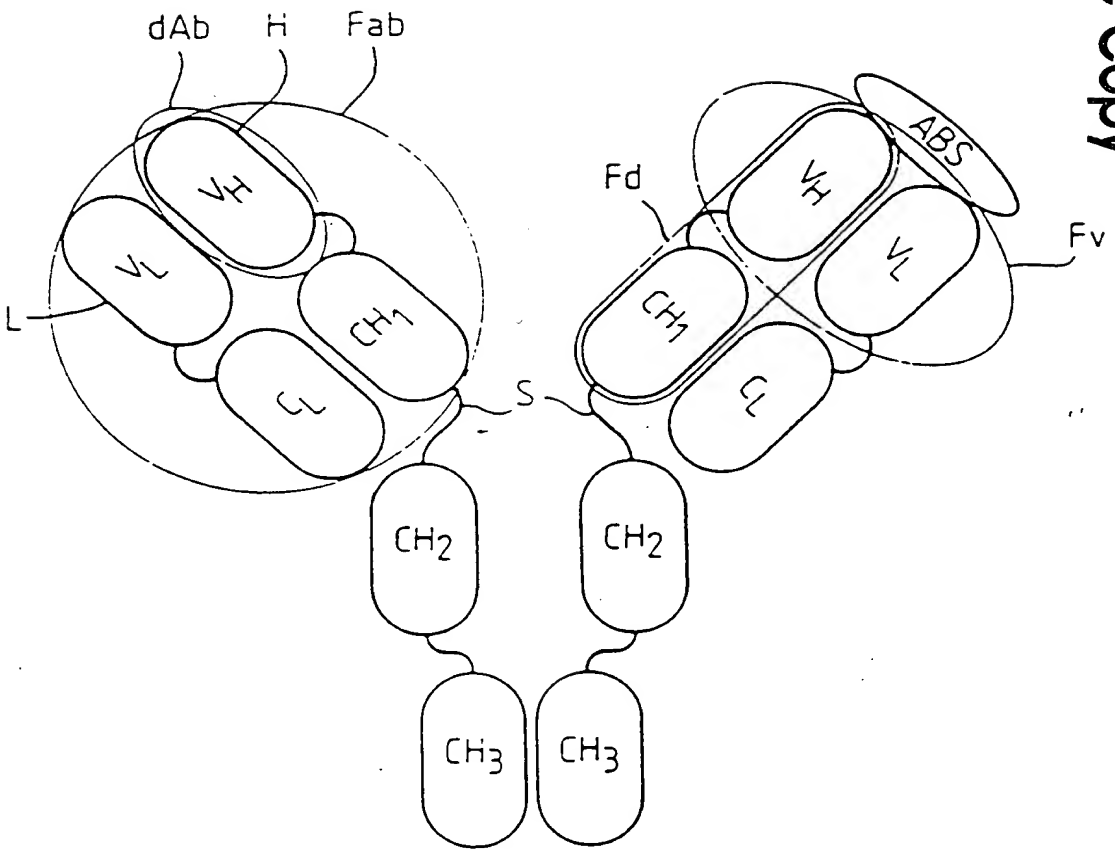


Fig. 2 (i)

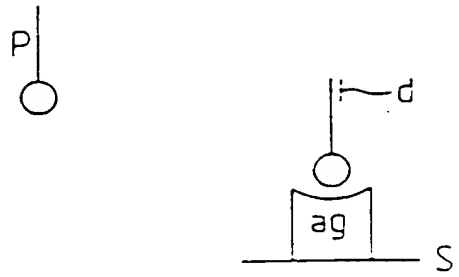
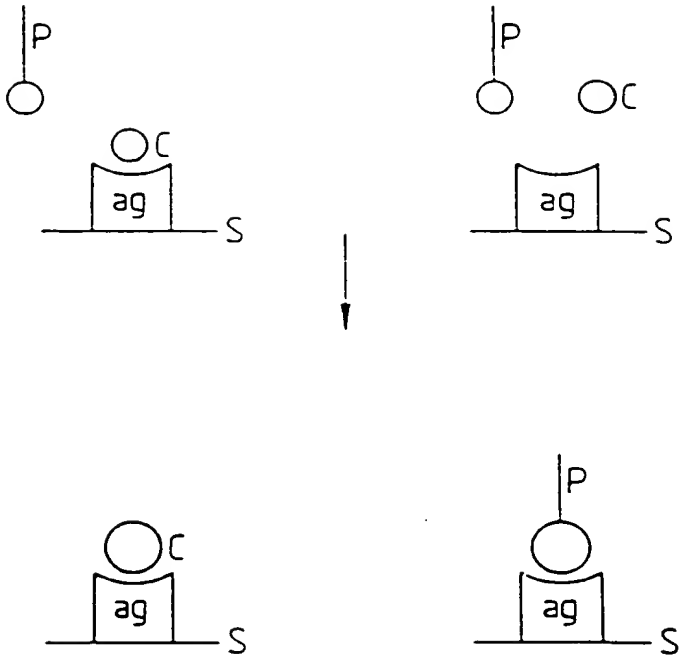


Fig. 2(ii)



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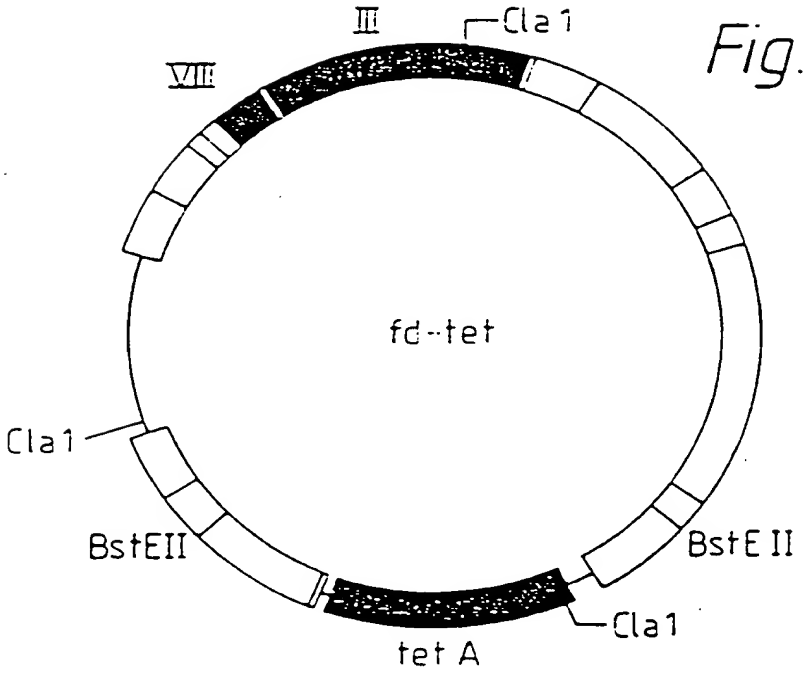

Fig. 3.

fd - tet

:

cleave with BstEII

:

fill in with Klenow

:

re-ligate

!

FDT δ Bst

:

in vitro mutagenesis (oligo 1)

!

FDTPs/Bs

:

in vitro mutagenesis (oligo 2)

!

FDTPs/Xh

Fig. 4.1

(1653)

Oligo 1 ACA ACT TTC AAC AGT TGA GGA GAC GGT GAC CGT AAG CTT CTG CAG TTG GAC CTG AGC GGA GTG AGA ATA (1620)

(1653)

Oligo 2 ACA ACT TTC AAC AGT TTC CCG TTT GAT CTC GAG CTC CTG CAG TTG GAC CTG

(1704)

Oligo 3 GTC GTC TTT CCA GAC GTT AGT

Fig. 4.2

GENE III

SIGNAL CLEAVAGE SITE

GENE III

(1624)

A TCT CAC TCC GCT

(1650)

GAA ACTGTT GAA AGT

Q V Q L Q V T V S S

B TCT CAC TCC GCT CAG GTC CAA CTG CAG AAG CTT ACG GTC ACC GTC TCC TCA ACT GTT GAA AGT

PstI BstEII

Q V Q L Q L E I K R

C TCT CAC TCC GCT CAG GTC CAA CTG CAG GAG CTC GAG ATC AAA CGG GAA ACT GTT GAA AGT

PstI XhoI

Fig. 5.

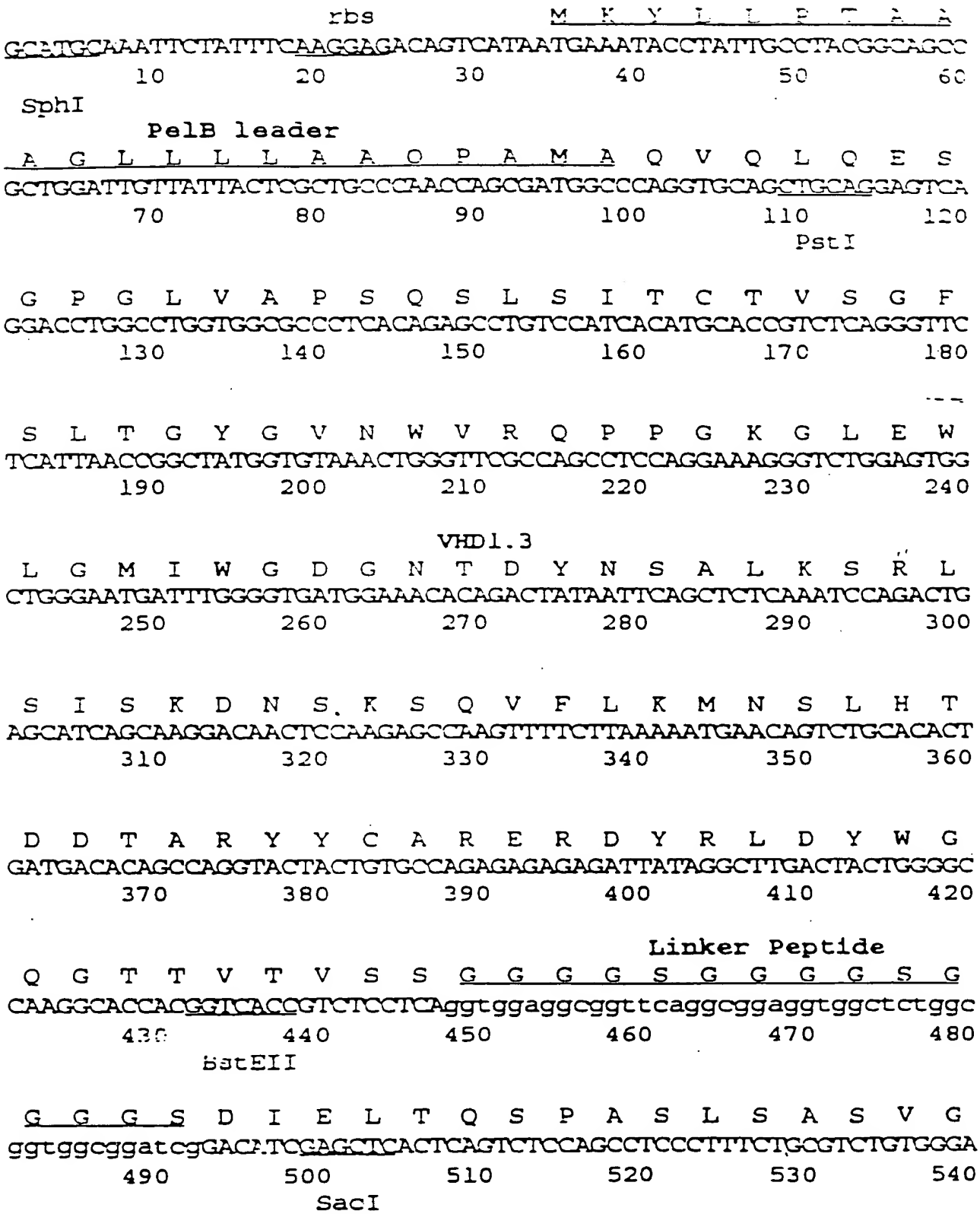
rbs
M K Y L L P T A A
GCATGCAAATTCTATTTCAAGGAGACAGTCATAATGAAATACCTATTGCCTACGGCAGCC
10 20 30 40 50 60
SphI
PelB leader
A G L L L L A A Q P A M A Q V Q L Q E S
GCTGGATTGTTATTACTCGCTGCCCAACCAGCGATGGCCCAGGTGCAGCTGCAGGAGTCA
70 80 90 100 110 120
PstI
G P G L V A P S Q S L S I T C T V S G F
GGACCTGGCCTGGTGGCGCCCTCACAGAGCCTGTCCATCACATGCACCGTCTCAGGGTTC
130 140 150 160 170 180
S L T G Y G V N W V R Q P P G K G L E W
TCATTAACCGGCTATGGTGTAAACTGGGTTCGCCAGCCTCCAGGAAAGGGTCTGGAGTGG
190 200 210 220 230 240
VHD1.3
L G M I W G D G N T D Y N S A L K S R L
CTGGGAATGATTTGGGGTGATGGAAACACAGACTATAATTCAGCTCTCAAATCCAGACTG
250 260 270 280 290 300
S I S K D N S K S Q V F L K M N S L H T
AGCATCAGCAAGGACAACTCCAAGAGCCAAGTTTTCTTAAAAATGAACAGTCTGCACACT
310 320 330 340 350 360
D D T A R Y Y C A R E R D Y R L D Y W G
GATGACACAGCCAGGTACTACTGTGCCAGAGAGAGAGATTATAGGCTTGACTACTGGGGC
370 380 390 400 410 420
Linker Peptide
Q G T T V T V S S G G G G S G G G G S G
CAAGGCACCACGGTCACCGTCTCCTCAggtggaggcggttcaggcggaggtggctctggc
430 440 450 460 470 480
BstEII
G G G S D I E L T Q S P A S L S A S V G
ggtggcggatcgGACATCGAGCTCACTCAGTCTCCAGCCTCCCTTTCTGCGTCTGTGGGA
490 500 510 520 530 540
SacI

Fig. 5 cont.

```
 E  T  V  T  I  T  C  R  A  S  G  N  I  H  N  Y  L  A  W  Y
GAAACTGTCACCATCACATGTCGAGCAAGTGGGAATATTCACAATTATTTAGCATGGTAT
      550        560        570        580        590        600

 Q  Q  K  Q  G  K  S  P  Q  L  L  V  Y  Y  T  T  T  L  A  D
CAGCAGAAACAGGGAAAATCTCCTCAGCTCCTGGTCTATTATACAACAACCTTAGCAGAT
      610        620        630        640        650        660

                                VKD1.3
 G  V  P  S  R  F  S  G  S  G  S  G  T  Q  Y  S  L  K  I  N
GGTGTGCCATCAAGGTTCAGTGGCAGTGGATCAGGAACACAATATTCTCTCAAGATCAAC
      670        680        690        700        710        720

 S  L  Q  P  E  D  F  G  S  Y  Y  C  Q  H  F  W  S  T  P  R
AGCCTGCAACCTGAAGATTTTGGGAGTTATTACTGTCAACATTTTTGGAGTACTCCTCGG
      730        740        750        760        770        780

                                                Myc Tag (TAG1)
 T  F  G  G  G  T  K  L  E  I  K  R  E  Q  K  L  I  S  E  E
ACGTTCGGTGGAGGGACCAAGCTCGAGATCAAACGGGAACAAAAACTCATCTCAGAAGAG
      790        800        810        820        830        840
                   XhoI

 D  L  N  *  *
GATCTGAATTAATAATGATCAAACGGTAATAAGGATCCAGCTCGAATTC
      850        860        870        880
                                              EcoRI
```

Fig. 6.

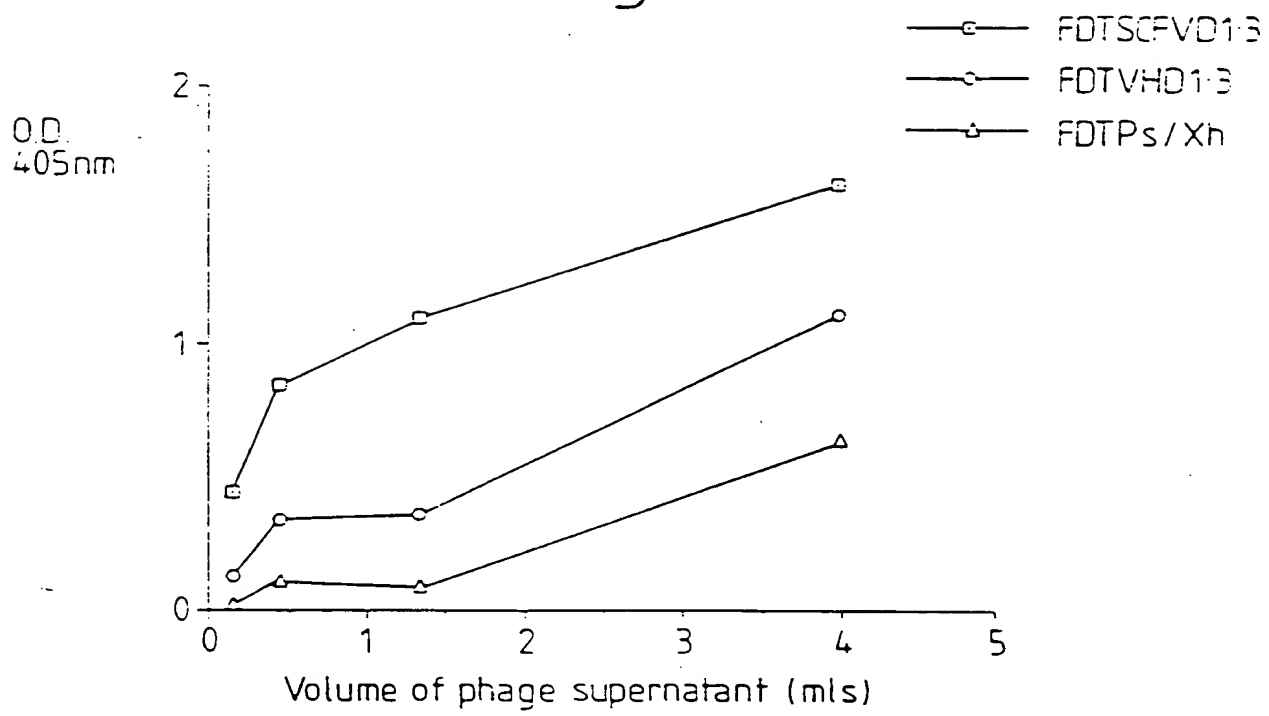

Fig. 7.

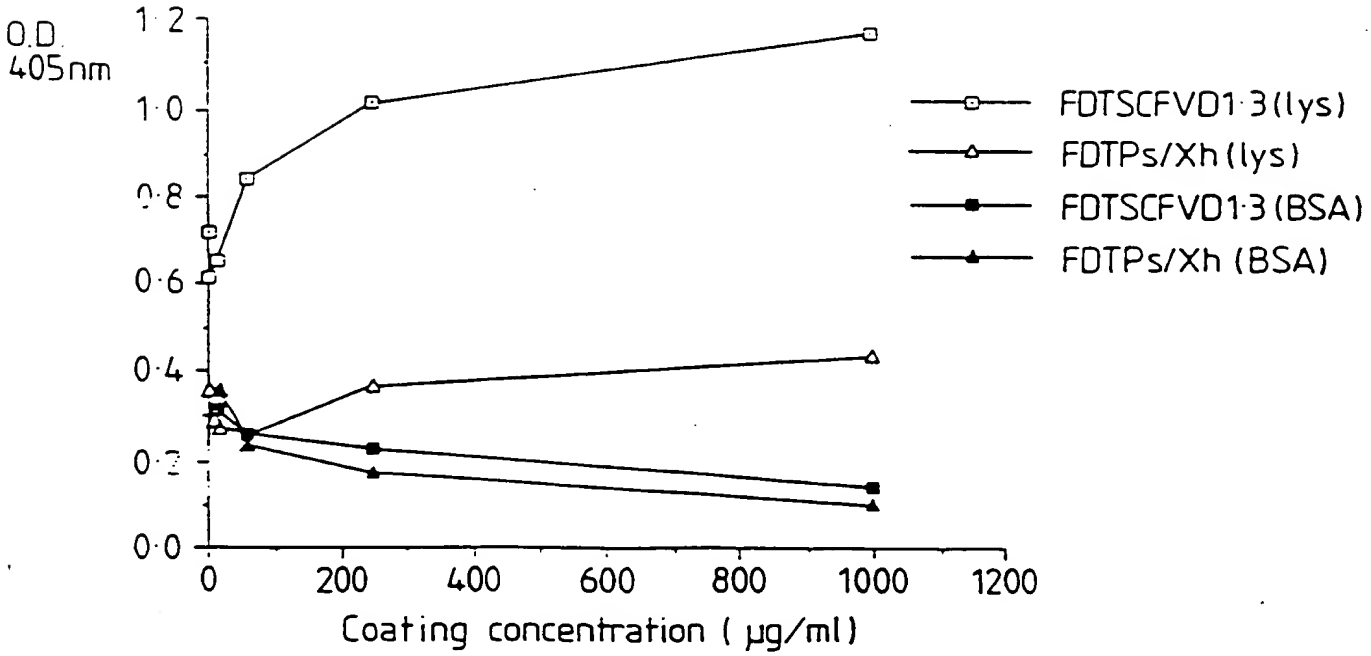

Fig. 8.

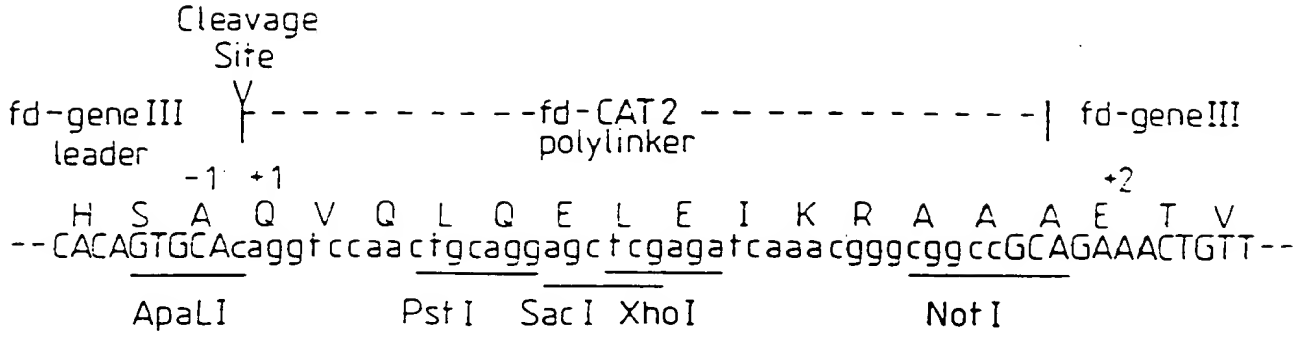
Cleavage Site
fd-gene III leader
fd-CAT2 polylinker
fd-gene III
-1 +1
+2
H S A Q V Q L Q E L E I K R A A A E T V
--CACAGTGCAcaggtccaactgcaggagctcgagatcaaacgggcggccGCAGAAACTGTT--
ApaLI
PstI SacI XhoI
NotI

Fig. 9.

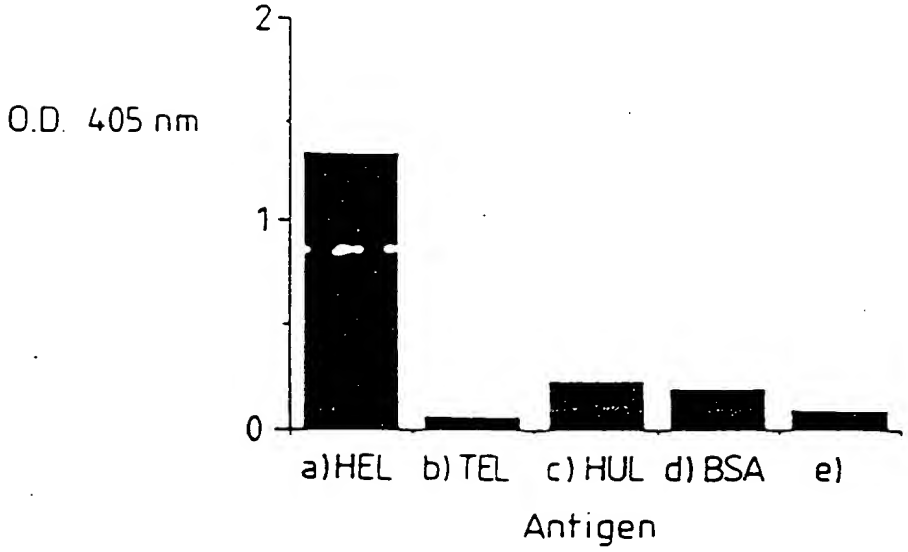
O.D. 405 nm
2
1
0
a) HEL
b) TEL
c) HUL
d) BSA
e)
Antigen

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Fig. 10.

```
                                                                M  K  Y  L  L  P  T  A  A
GCATGCAAATTCTATTTCAAGGAGACAGTCATAATGAAATACCTATTGCCTACGGCAGCC
        10        20        30        40        50        60

 A  G  L  L  L  L  A  A  Q  P  A  M  A  Q  V  Q  L  Q  E  S
GCTGGATTGTTATTACTCGCTGCCCAACCAGCGATGGCCCAGGTGCAGCTGCAGGAGTCA
        70        80        90       100       110       120

 G  P  G  L  V  A  P  S  Q  S  L  S  I  T  C  T  V  S  G  F
GGACCTGGCCTGGTGGCGCCCTCACAGAGCCTGTCCATCACATGCACCGTCTCAGGGTTC
       130       140       150       160       170       180

 S  L  T  G  Y  G  V  N  W  V  R  Q  P  P  G  K  G  L  E  W
TCATTAACCGGCTATGGTGTAAACTGGGTTCGCCAGCCTCCAGGAAAGGGTCTGGAGTGG
       190       200       210       220       230       240

 L  G  M  I  W  G  D  G  N  T  D  Y  N  S  A  L  K  S  R  L
CTGGGAATGATTTGGGGTGATGGAAACACAGACTATAATTCAGCTCTCAAATCCAGACTG
       250       260       270       280       290       300

 S  I  S  K  D  N  S  K  S  Q  V  F  L  K  M  N  S  L  H  T
AGCATCAGCAAGGACAACTCCAAGAGCCAAGTTTTCTTAAAAATGAACAGTCTGCACACT
       310       320       330       340       350       360

 D  D  T  A  R  Y  Y  C  A  R  E  R  D  Y  R  L  D  Y  W  G
GATGACACAGCCAGGTACTACTGTGCCAGAGAGAGATTATAGGCTTGACTACTGGGGC
       370       380       390       400       410       420

 Q  G  T  T  V  T  V  S  S  A  S  T  K  G  P  S  V  F  P  L
CAAGGCACCACGGTCACCGTCTCCTCAGCCTCCACCAAGGGCCCATCGGTCTTCCCCCTG
       430       440       450       460       470       480

 A  P  S  S  K  S  T  S  G  G  T  A  A  L  G  C  L  V  K  D
GCACCCTCCTCCAAGAGCACCTCTGGGGGCACAGCGGCCCTGGGCTGCCTGGTCAAGGAC
       490       500       510       520       530       540
```

Fig.10 cont.(1)

```
 Y  F  P  E  P  V  T  V  S  W  N  S  G  A  L  T  S  G  V  H
TACTTCCCCGAACCGGTGACGGTGTCGTGGAACTCAGGCGCCCTGACCAGCGGCGTGCAC
      550        560        570        580        590        600

 T  F  P  A  V  L  Q  S  S  G  L  Y  S  L  S  S  V  V  T  V
ACCTTCCCGGCTGTCCTACAGTCCTCAGGACTCTACTCCCTCAGCAGCGTGGTGACCGTG
      610        620        630        640        650        660

 P  S  S  S  L  G  T  Q  T  Y  I  C  N  V  N  H  K  P  S  N
CCCTCCAGCAGCTTGGGCACCCAGACCTACATCTGCAACGTGAATCACAAGCCCAGCAAC
      670        680        690        700        710        720

 T  K  V  D  K  K  V  E  P  K  S  S  *  *
ACCAAGGTCGACAAGAAAGTTGAGCCCAAATCTTCATAATAACCCGGGAGCTTGCATGCA
      730        740        750        760        770        780

                                 M  K  Y  L  L  P  T  A  A  A  G  L
AATTCTATTTCAAGGAGACAGTCATAATGAAATACCTATTGCCTACGGCAGCCGCTGGAT
      790        800        810        820        830        840

 L  L  L  A  A  Q  P  A  M  A  D  I  E  L  T  Q  S  P  A  S
TGTTATTACTGCTGCCCAACCAGCCATGGCCGACATCGAGCTCACCCAGTCTCCAGCCT
      850        860        870        880        890        900

 L  S  A  S  V  G  E  T  V  T  I  T  C  R  A  S  G  N  I  H
CCCTTTCTGCGTCTGTGGGAGAAACTGTCACCATCACATGTCGAGCAAGTGGGAATATTC
      910        920        930        940        950        960

 N  Y  L  A  W  Y  Q  Q  K  Q  G  K  S  P  Q  L  L  V  Y  Y
ACAATTATTTAGCATGGTATCAGCAGAAACAGGGAAAATCTCCTCAGCTCCTGGTCTATT
      970        980        990       1000       1010       1020
```

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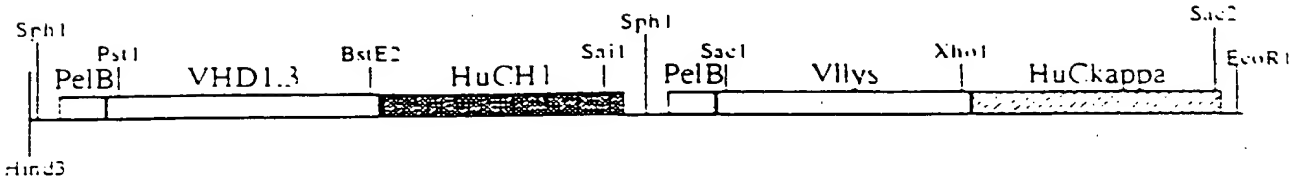
Fig. 10 cont. (2)

```
   T  T  T  L  A  D  G  V  P  S  R  F  S  G  S  G  S  G  T  C
ATACAACAACCTTAGCAGATGGTGTGCCATCAAGGTTCAGTGGCAGTGGATCAGGAACAC
      1030       1040       1050       1060       1070       1080

   Y  S  L  K  I  N  S  L  Q  P  E  D  F  G  S  Y  Y  C  Q  H
AATATTCTCTCAAGATCAACAGCCTGCAGCCTGAAGATTTTGGGAGTTATTACTGTCAAC
      1090       1100       1110       1120       1130       1140

   F  W  S  T  P  R  T  F  G  G  G  T  K  L  E  I  K  R  T  V
ATTTTTGGAGTACTCCTCGGACGTTCGGTGGAGGCACCAAGCTCGAGATCAAACGGACTG
      1150       1160       1170       1180       1190       1200

   A  A  P  S  V  F  I  F  P  P  S  D  E  Q  L  K  S  G  T  A
TGGCTGCACCATCTGTCTTCATCTTCCCGCCATCTGATGAGCAGTTGAAATCTGGAACTG
      1210       1220       1230       1240       1250       1260

   S  V  V  C  L  L  N  N  F  Y  P  R  E  A  K  V  Q  W  K  V
CCTCTGTTGTGTGCCTGCTGAATAACTTCTATCCCAGAGAGGCCAAAGTACAGTGGAAGG
      1270       1280       1290       1300       1310       1320

   D  N  A  L  Q  S  G  N  S  Q  E  S  V  T  E  Q  D  S  K  D
TGGATAACGCCCTCCAATCGGGTAACTCCCAGGAGAGTGTCACAGAGCAGGACAGCAAGG
      1330       1340       1350       1360       1370       1380

   S  T  Y  S  L  S  S  T  L  T  L  S  K  A  D  Y  E  K  H  K
ACAGCACCTACAGCCTCAGCAGCACCCTGACGCTGAGCAAAGCAGACTACGAGAAACACA
      1390       1400       1410       1420       1430       1440

   V  Y  A  C  E  V  T  H  Q  G  L  S  S  P  V  T  K  S  F  N
AAGTCTACGCCTGCGAAGTCACCCATCAGGGCCTGAGCTCGCCCGTCACAAAGAGCTTCA
      1450       1460       1470       1480       1490       1500

   R  G  E  S  *  *
ACCGCGGAGAGTCATAGTAAGAATTC
      1510       1520
```

Fig. 10 cont. (3)



FabD1.3 in pUC19

Fig. 11.

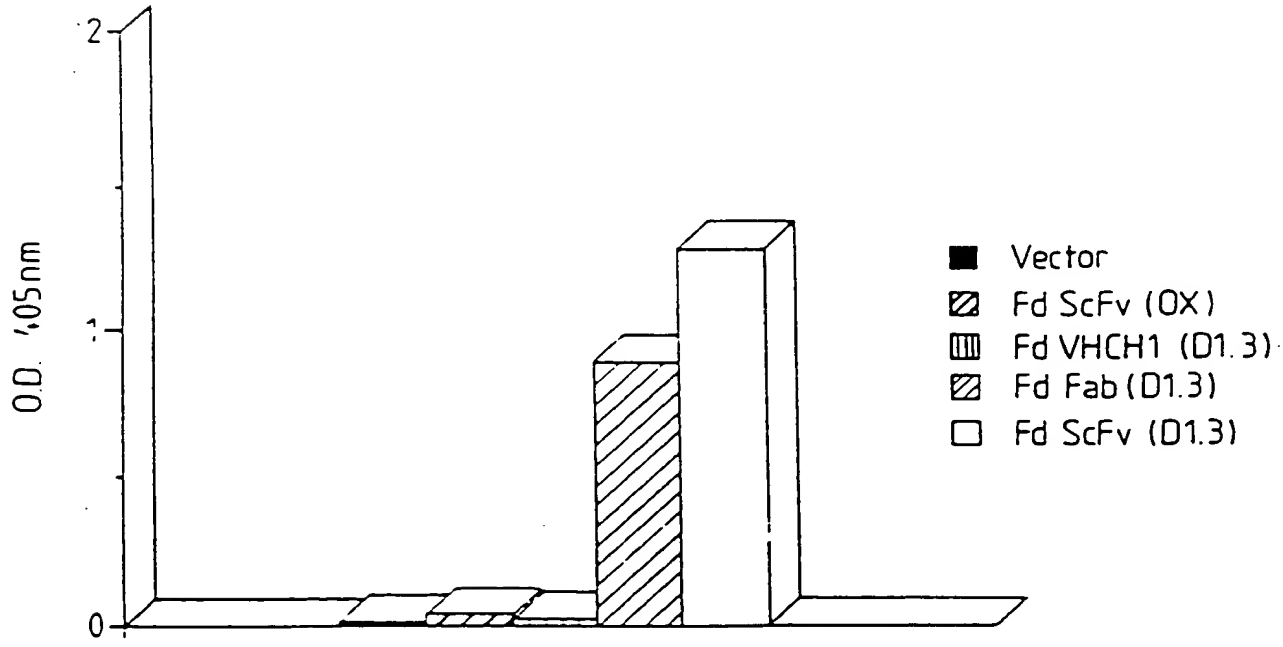


Fig. 12.

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Fig. 13.

```
 Q   V   Q   L   Q   E   S   G   G   G   L   V   Q   P   G   G
CAG GTG CAG CTG CAG GAG TCA GGA GGA GGC TTG GTA CAG CCT GGG GGT
              PstI
 S   L   R   L   S   C   A   T   S   G   F   T   F   S   N   Y
TCT CTG AGA CTC TCC TGT GCA ACT TCT GGG TTC ACC TTC AGT AAT TAC

 Y   M   G   W   V   R   Q   P   P   G   K   A   L   E   W   L
TAC ATG GGC TGG GTC CGC CAG CCT CCA GGA AAG GCA CTT GAG TGG TTG

 G   S   V   R   N   K   V   N   G   Y   T   T   E   Y   S   A
GGT TCT GTT AGA AAC AAA GTT AAT GGT TAC ACA ACA GAG TAC AGT GCA

 S   V   K   G   R   F   T   I   S   R   D   N   F   Q   S   I
TCT GTG AAG GGG CGG TTC ACC ATC TCC AGA GAT AAT TTC CAA AGC ATC

 L   Y   L   Q   I   N   T   L   R   T   E   D   S   A   T   Y
CTC TAT CTT CAA ATA AAC ACC CTG AGA ACT GAG GAC AGT GCC ACT TAT

 Y   C   A   R   G   Y   D   Y   G   A   W   F   A   Y   W   G
TAC TGT GCA AGA GGC TAT GAT TAC GGG GCC TGG TTT GCT TAC TGG GGC

 Q   G   T       L   V   T   v   s   s   g g g g s g g g g s
CAA GGG ACC   CTG GTC ACC gtc tcc tca ggtggaggcggttcaggcggaggtggctct
                  BstEII
 g g g g s d   i   E   L   T   Q   T   P   L   S   L   P   V
ggcggtggcggctcggac atc GAG CTC ACC CAA ACT CCA CTC TCC CTG CCT GTC
                       SacI
 S   L   G   D   Q   A   S   I   S   C   R   S   S   Q   S   I
AGT CTT GGA GAT CAA GCC TCC ATC TCT TGC AGA TCT AGT CAG AGC ATT

 V   H   S   N   G   N   T   Y   L   E   W   Y   L   Q   K   P
GTA CAT AGT AAT GGA AAC ACC TAT TTA GAA TGG TAC CTG CAG AAA CCA
                                            PstI
 G   Q   S   P   K   L   L   I   Y   K   V   S   N   R   F   S
GGC CAG TCT CCA AAG CTC CTG ATC TAC AAA GTT TCC AAC CGA TTT TCT

 G   V   P   D   R   F   S   G   S   G   S   G   T   D   F   T
GGG GTC CCA GAC AGG TTC AGT GGC AGT GGA TCG GGG ACA GAT TTC ACA

 L   K   I   S   R   V   E   A   E   D   L   G   V   Y   Y   C
CTC AAG ATC AGC AGA GTG GAG GCT GAG GAT CTG GGA GTT TAT TAC TGC

 F   Q   G   S   H   V   P   Y   T   F   G   G   G   T   K   L
TTT CAA GGT TCA CAT GTT CCG TAC ACG TTC GGA GGG GGG ACC AAG CTC

 E   I   K   R
GAG ATC AAA CGG
XhoI
```

Fig. 14.

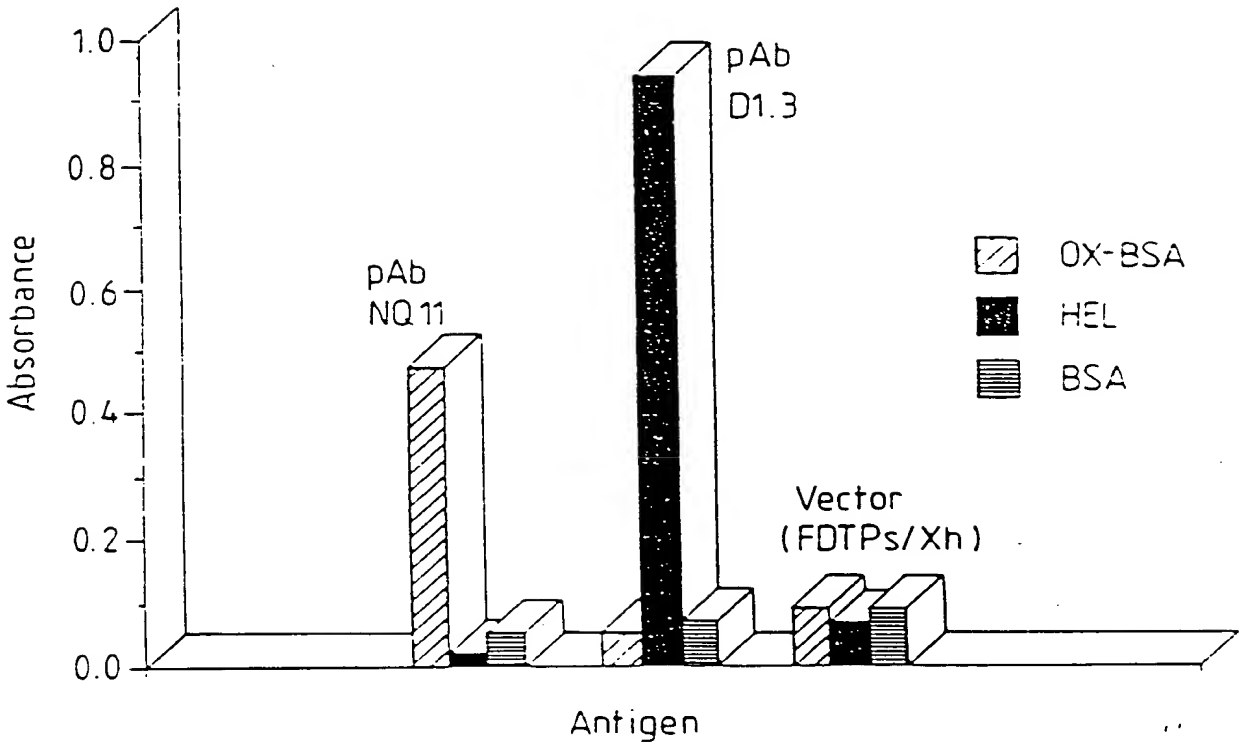

Fig. 15.

5' END

R T P E M P V L

TCT CAC AGT GCA CAA ACT GTT GAA CGG ACA CCA GAA ATG CCT GTT CTG

ApaL1

3' END

K A A L G L K

AAA GCC GCT CTG GGG CTG AAA GCG GCC GCA GAA ACT GTT GAA AGT etc.

Not I

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Fig. 16(1)

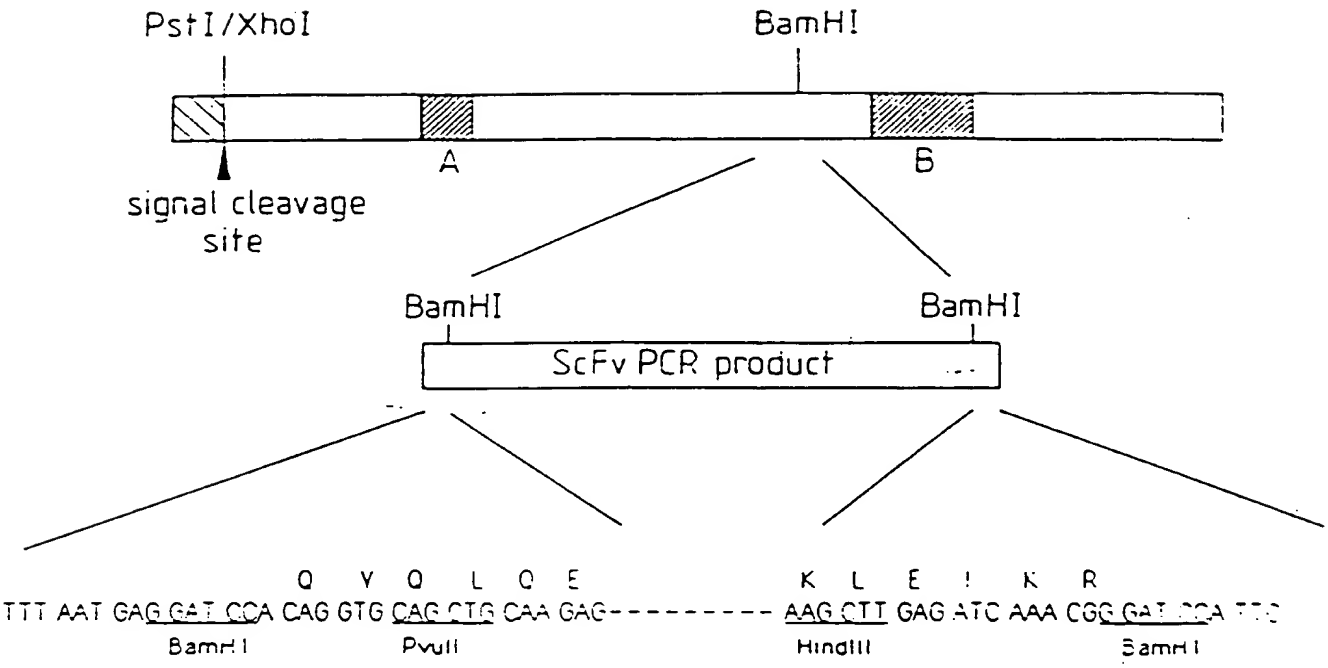


Fig. 16(2)

A (1834) 5' GAG GGT GGT GGC TCT
" " "C " "
" " "C " "
" " "C " ACT 3'(1839)

B (2284) 5' - GGC GGC GGC TCT
- GGT GGT GGT "
- " GGC GGC "
GAG " " GGC "
" " " GGT "
" " " GGC "
" " " GGT "
- " " GGC " 3'(2379)

Reverse complement of mutagenic oligo G3Bamlink

T
5' GAG GGT GGC GGA TCC

T
GAG GGT GGC GG 3'

Fig.17.

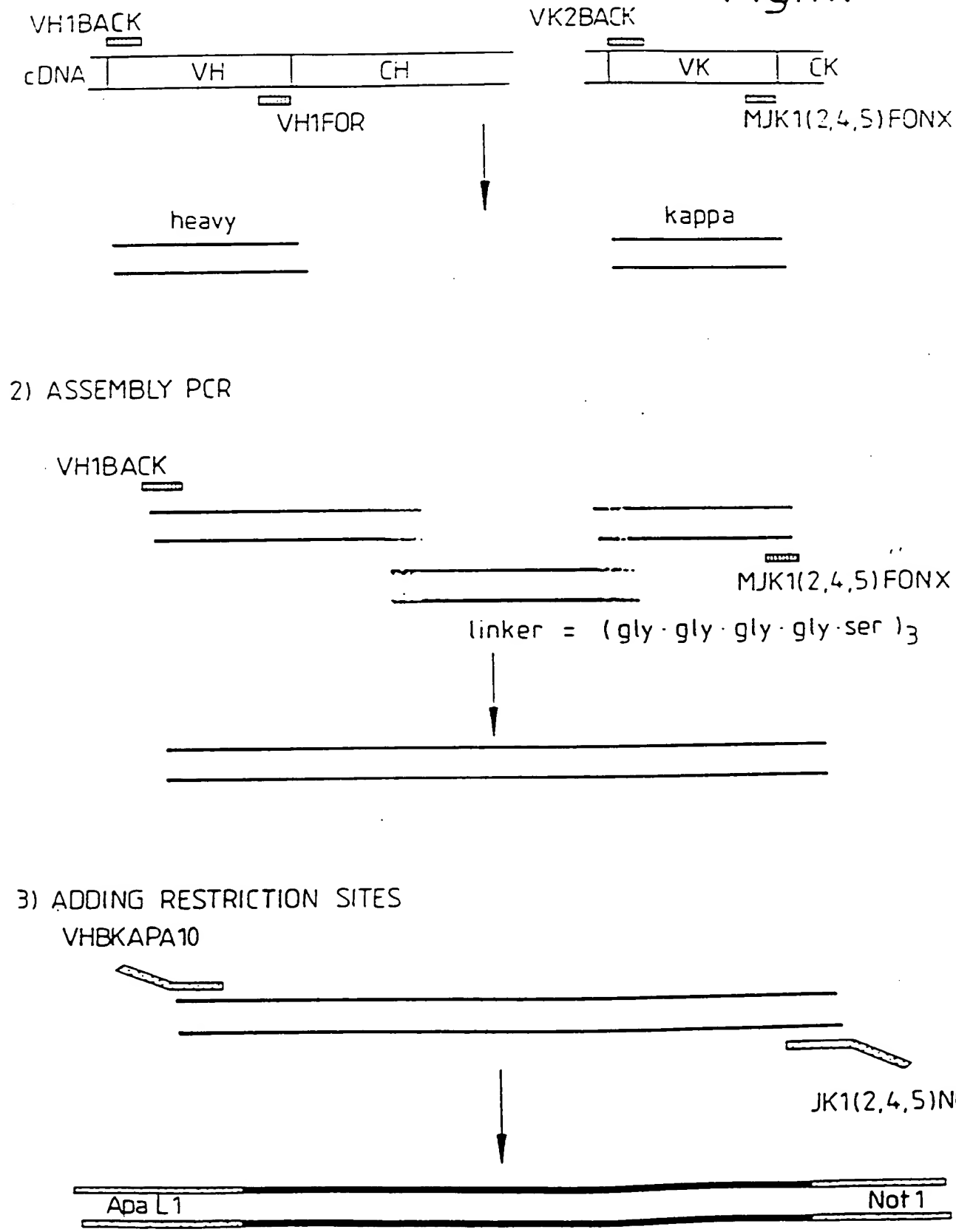


Fig. 18.

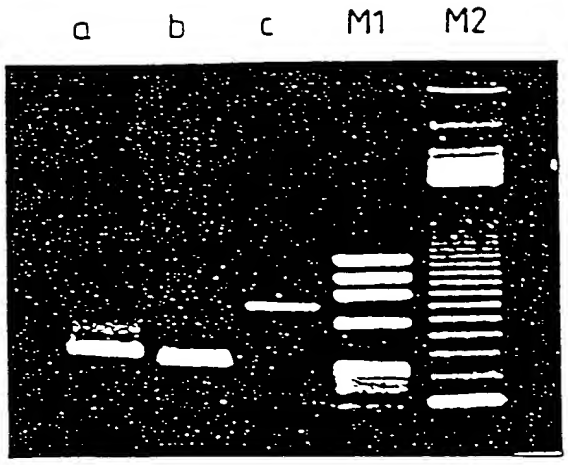


Fig. 19.

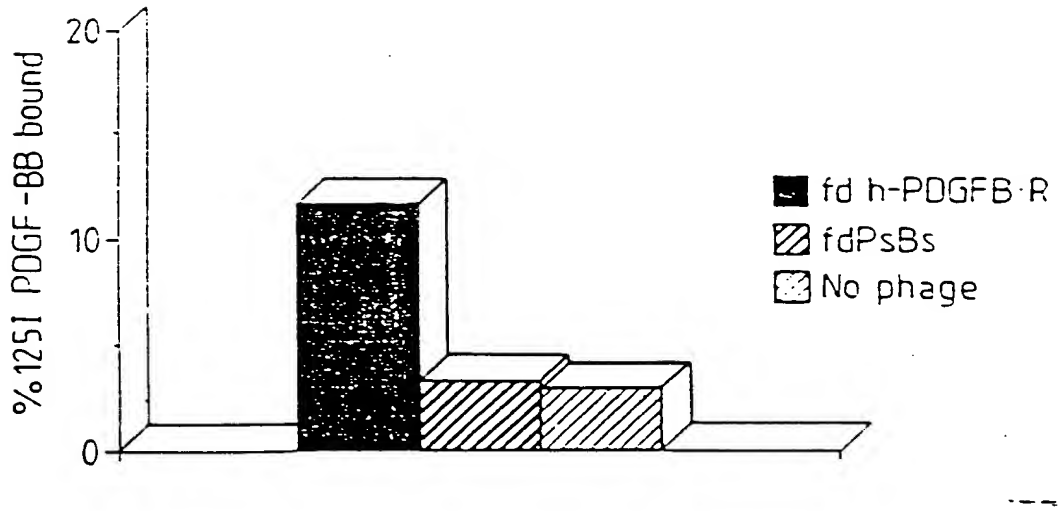


Fig. 20.

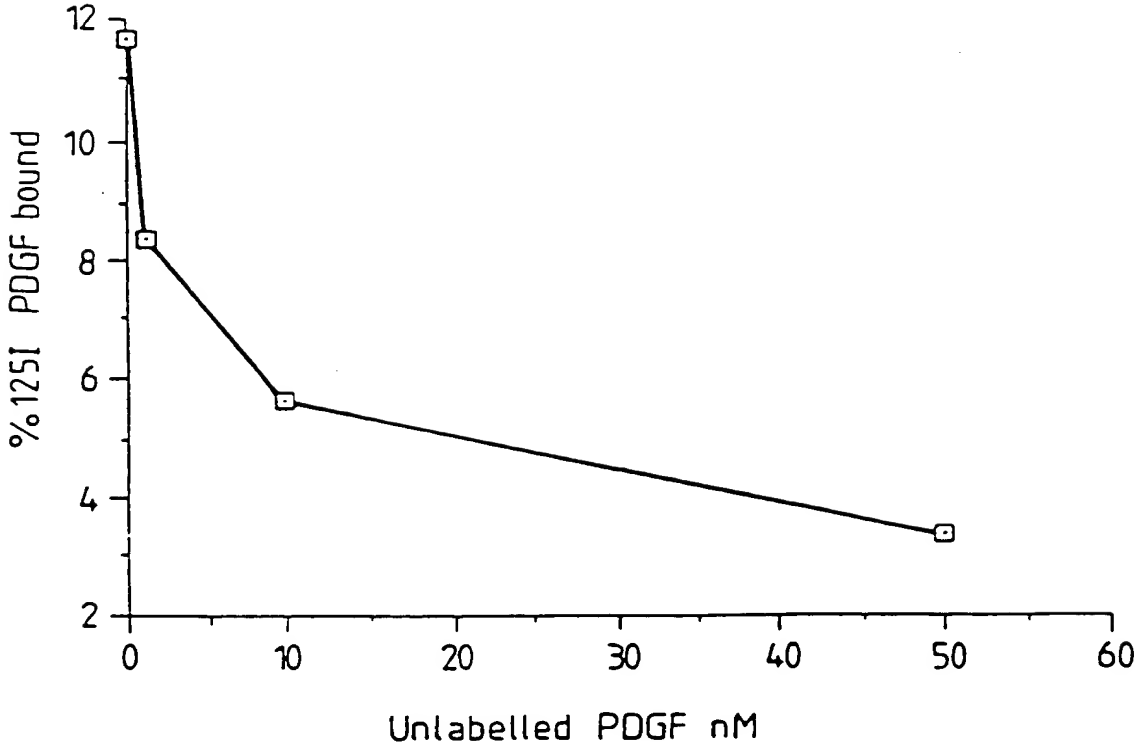


Fig. 21.

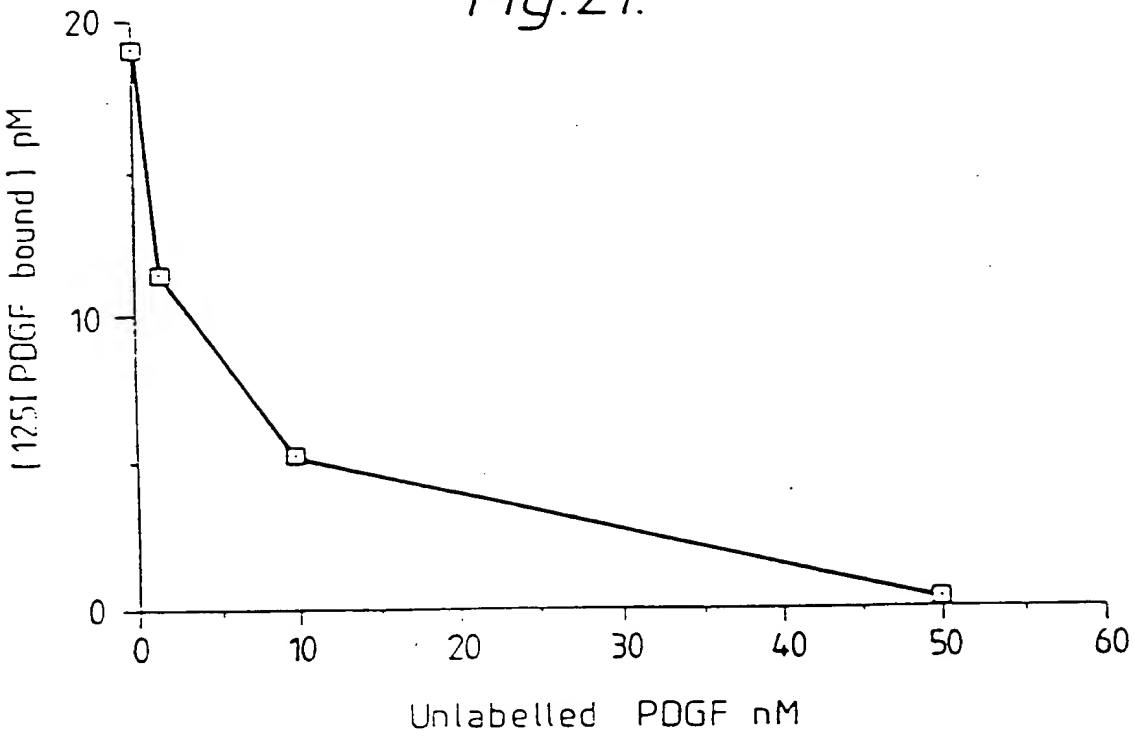


Fig. 22.

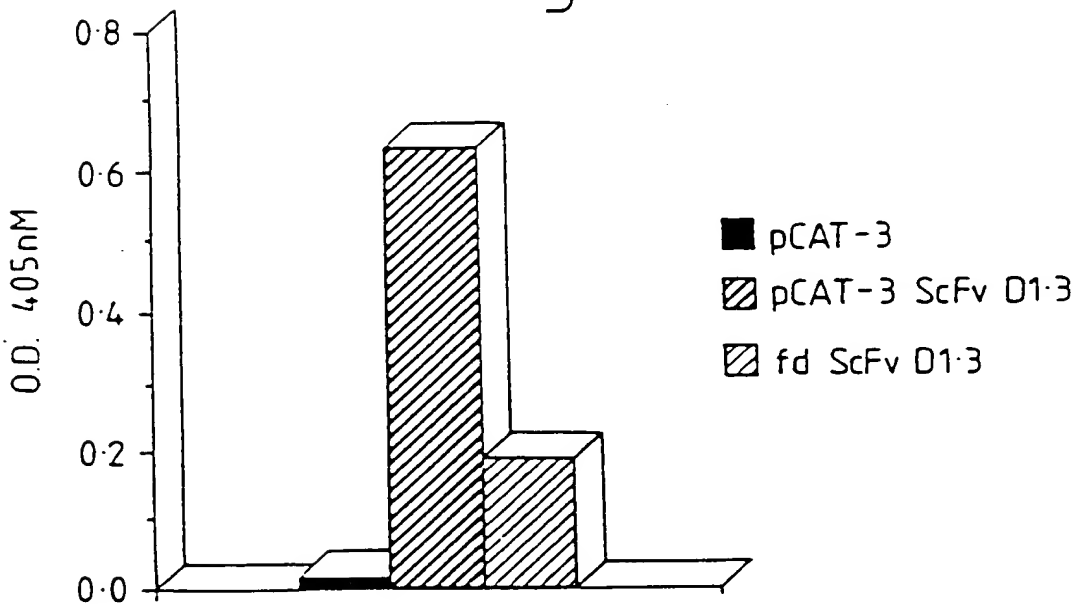
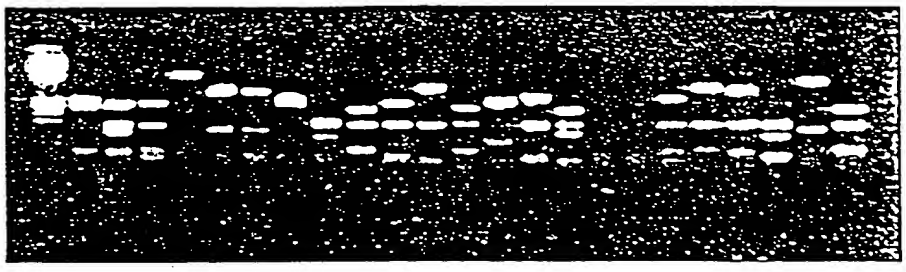


Fig. 23.

d

M



M

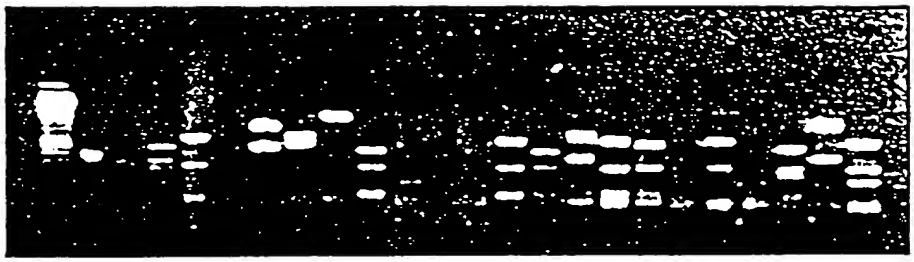


Fig. 24.

VH sequences

from combinatorial library:

	CDR1		CDR2		CDR3		
A	QVQLQQSGAELARPGASVKMSCKASGYTFT	SYTMQI	WVKQRPGQGLEWIG	YINPSSGYTNYNQKFKD	KATLTADKSSSTAYMQLSSLTSEDSAVYYCAII	RYGAY	WGQGTTVTVSA x4
B	QVKLQQSGAELARPGASVKMSCKASGYTFT	RDMHQI	WLKQRPGQGLEWIG	YIIPSTGYTEYNQKFKD	KATLTADKSSSTAYMQLSSLTSEDSAVYYCAR	IIYGLY	WGQGTTVTVSS x9
C	QVQLQQSGAELARPGASVKMSCKASGYTFT	SYVMQI	WVKQRPGQGLEWIG	YIIPYNGDTFYNQKFKG	KATLTVDKSSSTAIMELSSLTSEDSAVYYCVG	YRSFPY	WGQGTTVTVSS x3
D	QVQLQQSGPELVKPGASVKISCKASGYSFT	SYVMN	WVKQSHGKSLEWIG	RIIPYNGDTFYNQKFKG	RLSITKDNSKSQVFLKMNSLQTDDTAMYYCAR	ITTRFAY	WGQGTTVTVSS x1
E	QVQLQESGPGLVAPSQSLSITCTVSGFSLT	SYGVH	WVRQPPGKGLEWLG	VIWAGGSTNYNSALMS	KATLTADKSSSTAYMQLSSLTSEDSAVYYCAR	DRGDY	WGQGTTVTVSS
F	QVQLQQSGPELAKPGASVKMSCKASGYTFT	RYLMQI	WVKQRPGQGLEWIG	YIIPSTGYTEYNQKFKD	KATLTADKSSSTAYMQLSSLTSEDSAVYYCAR	DYGYY	WGQGTTVTVSS
G	QVKLQQSGAELVRPGASVKLSCKASGYTFT	RIYMQI	WVKQRPGQGLEWIG	YIIPSTGYTEYNQKFKD	KATLTADKSSSTAYMQLSSLTSEDSAVYYCAT	DYGND	WGQGTTVTVSS
R	QVQLQQSGPELMKPGASVKISCKASGYSFS	RIYMQI	WVKQSHGKSLEWIG	YIAPFNGGTTYNQKFKG	KATLTVDRSSSTAYMQLSSLTSEDSAVYYCAT		

from hierarchical library VH-rep x Vκ-d:

I	QVKLQQSGPELARPGVSVKMSCKASGYTFT	SYAMQI	WVKQSQSKSLEWIG	VISTYNGNTNYNQKFKG	KATMTVDKSSSTAYMELARLTSEDSAIYYCAR	DYGDY	WGQGTTVTVSA
J	QVKLQQSGAELARPGASVKMSCKASGYTFT	RYTMQI	WVKQRPGQGLEWIG	YIIPSSGYTNYNQKFKD	KATLTADKSSSTAYMQLSSLTSEDSAVYYCAR	DRGAY	WGQGTTVTVSS
K	QVKLQQSGAELARPGASVKMSCKASGYTFT	RDMHQI	WVKQRPGQGLEWIG	YIIPSTGYTEYNQKFRD	KATLTADKSSSTAYMQLSSLTSEDSAVYYCAR	IIYGLY	WGQGTTVTVSS x3
L	QVQLQQSGLELARPGASVKMSCKASGYTFT	IIYMHI	WVKQRPGQGLEWIG	YIIPSTGYTEYNQKFKD	KATLTADKSSSTAYMQLSSLTSEDSAVYYCAR	DYGYY	WGQGTTVTVSS
N	QVKLQQSGAELAKPGASVKMSCKTSGYTFT	SYTMQI	WVKQRPGQGLEWIG	YIIPSSGYTNYNQKFKD	KATLTADKSSSTAYMQLSSLTSEDSAVYYCAR	DYGYF	WGQGTTVTVSS
O	QVQLQQSGAELVKPGASVKMSCEASGYTFT	SHLMQI	WVKQRPGQGLEWIG	YIIPSTGYTEYNQKFKD	KATLTADKSSSTAYMQLSSLTSEDSAVYYCAR	DYGYY	WGQGTTVTVSS
P	QVQLQQSGAELARPGASVKMSCKASGYTFT	SYVMQI	WVKQRPGQGLEWIG	YIIPSTGYTEYNQKFKD	KATLTADKSSSTAYMQLSSLTSEDSAVYYCAR	DYGAY	WGQGTTVTVSS
Q	QVKLQQSGAELARPGASVKMSCKASGYTFT	SYLMQI	WVKQRPGQGLEWIG	YIIPSTGYTEYNQKFKD	KATLTADKSSSTAYMQLSSLTSEDSAVYYCAR	DYGYY	WGQGTTVTVSS
R	QVKLQQSGAELAKPGASVKMSCKATGYTFT	SYVMQI	WVKQRPGQGLEWIG	YIIPSTGYTEYNQKFKD	KATLTADKSSSTAYMQLSSLTSEDSAVYYCAR	DYGYY	WGQGTTVTVSS
S	QVQLQQSGAELARPGASVKMSCKASGYTFT	TPLMQI	WLKQRPGQGLEWIG	YIIPSSGYTNYNQKFKD	KATLTADKSSSTAYMQLSSLTSEDSAVYYCAR	DYGYY	WGQGTTVTVSS
T	QVQLQQSGAELAKPGASVKMSCKASGYTFT	SYTMQI	WVKQRPGQGLEWIG	YIIPTTGYTEYNQKFKD	KATLTADKSSSTAYMQLSSLTSEDSAVYYCAR	DYGYY	WGQGTTVTVSS
U	QVKLQQSGAELARPGASVKMSCKASGYTFT	SYTMQI	WVKQRPGQGLEWIG	YIIPSTGYTEYNQKFKD	KATLTADKSSSTAYMQLSSLTSEDSAVYYCAR	DYGYY	WGQGTTVTVSS
B	QVKLQQSGAELAKPGASVKMSCKASGYTFT	RDMHQI	WLKQRPGQGLEWIG	YIIPSTGYTEYNQKFKD	KATLTADKSSSTAYMQLSSLTSEDSAVYYCAR	DYGYY	WGQGTTVTVSS

VH1x1

Fig. 24 cont.

Vκ sequences

from combinatorial library:

		CDR1		CDR2		CDR3				
a	DIELTQ8P88L.:A8LOERV8LTC	RA8QEI8OYL8	WLQQKPDO8IKRLIY	AA8TL88	OVPKRF0O8R8O0OY8LTI88LE080FADYYC	LQYA8YPT	FOAOTKLEIKRA	x1	V	ox·like
b	DIELTQ8PAIM8AnPOEKVTMTC	RA888V888YLH	WYQQK8OA8PKVWIY	8T8IILA8	OVPARFO0808OTTOY9LTI89VRAEDAATYYC	QQYnOYPLT	FOAOTKLEIKRA	x1	IV	ox·like
c	DIELTQ8PTTMAA8PO8KITITC	8A888I80IIYLH	WYQQKPOF8PKLLIY	RT8IILA8	OVPARFO080OKIT8Y8LTIOTM8AEDVATYYC	QQOnnIPLT	FOAOTKLEIKRA	x2	IV	ox·like
d	DIELTQ9PTTMAA8POEKITITC	8A888I88IIYLH	WFQQKPOF8PKLLI8	RT8IILA8	OVPARF90O0OOTT8Y0LTIOTMEAEDVATYYC	QQO8TIPPT	FO8OTKLEIKRA	x9	IV	ox·like
e	DIELTQ8PAIM8A8POEKVTITC	8A898VIIYMI	WFQQKPOT8PKLWIY	8T8IILA8	OVPTRFO0O0OTTOY0LTIORMEAEDAATYYC	QQRn8YPPT	FO8OTKLEIKRA	x4	VI	ox·like?
f	DIELTQ9PAIM8AFPOEKVTMTC	8A98RV8YMI	WYQQK8OT9PKRWIY	DT8KLA9	OVPARF80O0O0OT8Y0LTI80MEAEDAATYYC	QQFnmIPLT	FOAOTKLEIKRA		VI	Vκox1
g	DIELTQ9PAIM9A8POEKVTMTC	8A888IIIYMI	WYQQKPOA8PKRWIY	DT8KLA8	OVPARF8O6O90T8Y0I/TI89MEAEDAATYYC	HQRIIAYPWT	FOOOTKLEIKRA		VI	ox·like?

from hierarchical library VH-B x Vκ-rep:

		CDR1		CDR2		CDR3				
h	DIELTQ9PAIM8A8POEKVTMTC	8A888V8YMI	WYQQKOOT8PKRWIY	DT9KLA9	OVPARF8O0O8OT8Y8LTI89IEAEDAATYYC	QQW98IPLT	FOAOTKLEIKRA	x4	IV/VI	Vκox1
i	DIELTQ8PAIM8A8POEKVTITC	8A889V8YIH	WFQQKPOT8PKLWIY	8T9IILA8	OVPARF0O0O0OT8Y8LTIORMEAEDAATYYC	QQYH8YPLT	FOAOTKLEIKRA		V	ox·like?
j	DIELTQ8PTTMAA9POEKITITC	8A998I80IIYLH	WFQQKPOF8PKLLIY	RT8IILA8	OVPARF90808OT8Y8LTIOTMEAEDVATYYC	QQn88IPLT	FOOOTKLEIKRA		V	ox·like
k	DIELTQ9PTTMAA8PODHITITC	8AT88I98IIYLH	WYQQKPOF8PKLLIY	RT9IILA8	OVPPRF0O8O8OT8Y8LTIOAMEAEDVATYYC	QQOn8IPYT	FOAOTKLEIKRA		V	ox·like
l	DIELTQ8PTTMAA8PO8KITITC	8A888I88IIYLH	WYQQKPOF8PKLLIY	RT8IILA8	OVPARF8O8O8OT8Y8LTIOTMEAEDVATYYC	QQOn8IPYT	FOOOTKLEIKRA		V	ox·like
m	DIELTQ8PTTMAA8PO8KITITC	8A888I88IIHLH	WYQQKPOF8PKLLIY	RT9IILA8	OVPARF8O8O8OT8Y8LTIOTMEAEDVATYYC	QQ990IPYT	FOOOTKLEIKRA		V	ox·like
n	DIELTQ9PTTMAA8POEKITITC	SA898I89IIYLH	WYQQKPOF8PKLLIY	RT8IILA8	OVPARF90909OT9Y9LTIOTMEAEDVATYYC	QQ0981PFT	FOOOTKLEIKRA		V	ox·like
o	DIELTQ9PAIMAA8POEKITITC	SA888I88IIYLH	WYQQKPOF8FKLLIY	RT8IILA9	OVPARF9O8O8OT9Y8LTIOTMEAEDVATYYC	QQ088IPYT	FOOOTKLEIKRA	x2	V	ox·like
p	DIELTQ9PAIM9A8POEKVTMTC	8A988V8YMI	WYQQK8OT8PKRWIY	DT8KLA8	OVPARF8O6O9OT9Y8LTI69MEAEDVATYYC	QQW89IIPLT	FOAOTKLEIKRA	x2	IV/VI	Vκox1
q	DIELTQ8PAIM9A8PODKVTLTC	8A868VRYVN	WFQQK8OT8PKRWIY	DT9KLA8	OVPARP9O6O6OT0Y9LTI89MEAEDAATYYC	QQWT9IIPPT	FOOOTKLEIKRA		IV/VI	Vκox1
r	DIELTQ9PAIM8A9PORKVIMTC	8A988V8YMI	WYQQK8OT8PKRWIY	DT8KLA8	OVPARF8O6O9OTnY9LTI80MEAEDAATYYC	QQWnTIIALT	FOAOTKLEIKRA		IV/VI	Vκox1
s	DIELTQ9PAIM8A8POEKVTMTC	RA888VT08YLH	WYQQK9OA8FKLWVY	8T9IILA8	OVPARF8O9O9OT9Y8LTI89VEAEDAATYYC	QQYnOYPLT	FOAOTKLEIKRA		IV/VI	ox·like
t	DIELTQ8PAIM8A8POEKVTMTC	RA988V998YLH	WYQQK8OA8PKLWIY	8T9IILA8	OVPARFSO8O9OT8Y8LTI8RMEAEDAATYYC	QQRn9YPLT	FOAOTKLEIKRA		IV/VI	ox·like
u	DIELTQ9PAIM8A8POEKVTMTC	RA888V988YLH	WYQQK8OA8PKLWIY	8T8IILA8	OVPARF8O8O9OT8Y8LTI89VEAEDAATYYC	QQY8OYPLT	FOAOTKLEIKRA		IV/VI	ox·like
v	DIELTQ9PAIM8A8POEKVTMTC	RA898V998YLH	WFQQK0OA8PKLWIY	9T8IILP8	OVPARF9O9O9OT8Y8LTI88VEAEDAATYYC	QQYnOYPLT	FOOOTKLEIKRA		IV/VI	ox·like
c	DIELTQSPTTMAA9POEKITITC	9A999I59NYLH	WYQQKPOFSFKLLIY	RTSIILA9	GVPARFSOSO9GT9Y9LTIOTMEAEDVATYYC	QQOSSIPLT	FOAOTKLEIKRA	x1		

SUBSTITUTE SHEET

Fig. 25.

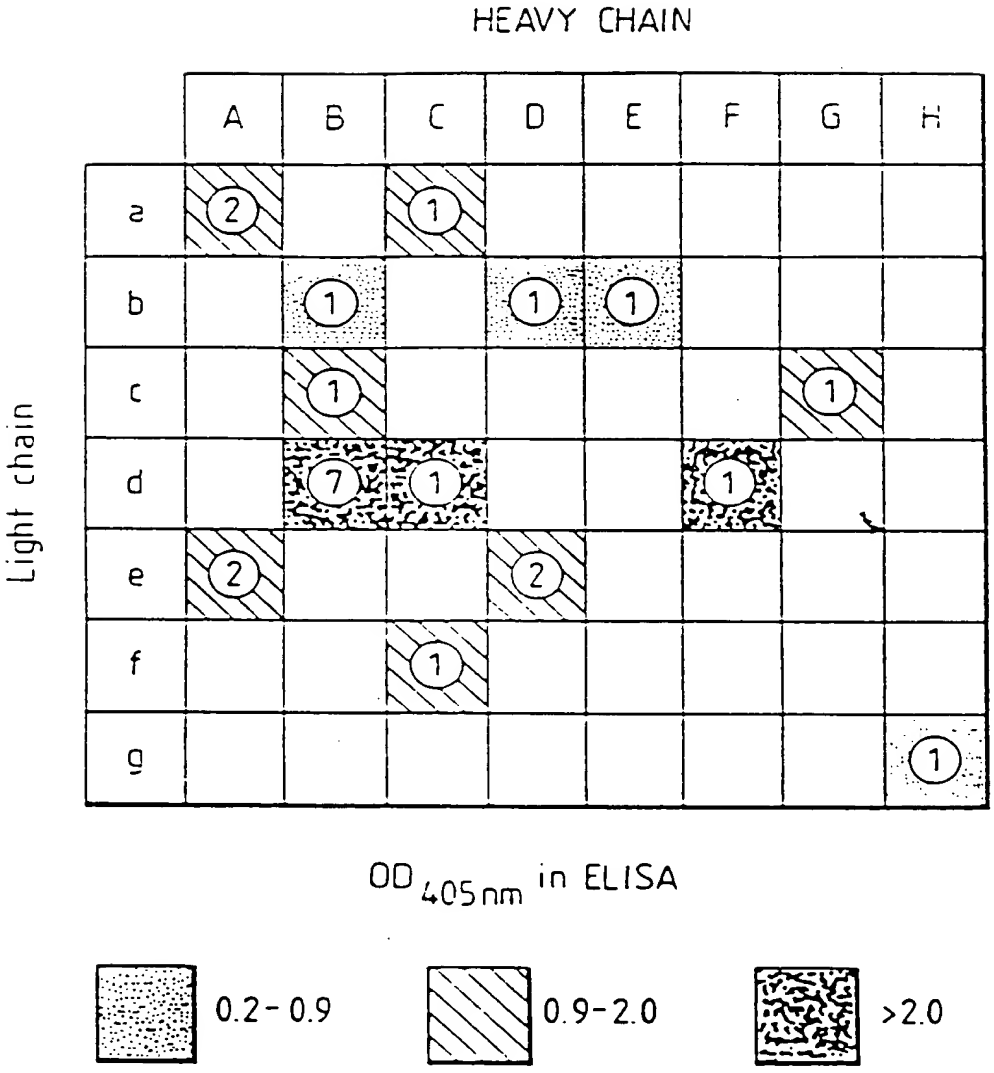

HEAVY CHAIN
A
B
C
D
E
F
G
H
a
b
c
d
e
f
g
Light chain
2
1
1
1
1
1
7
1
1
2
2
1
1
OD 405nm in ELISA
0.2-0.9
0.9-2.0
>2.0

Fig. 26(a)

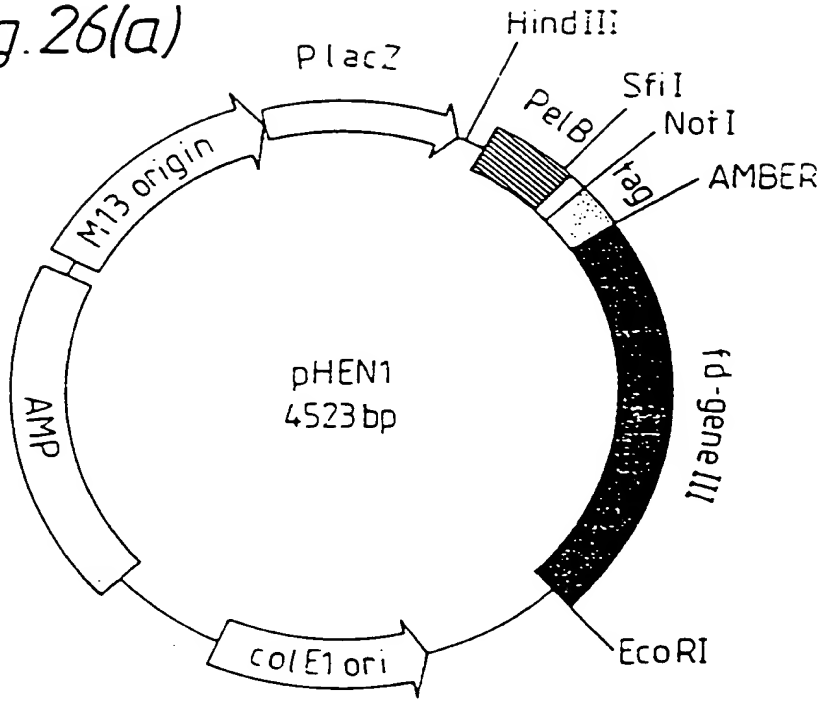


Fig. 26(b)

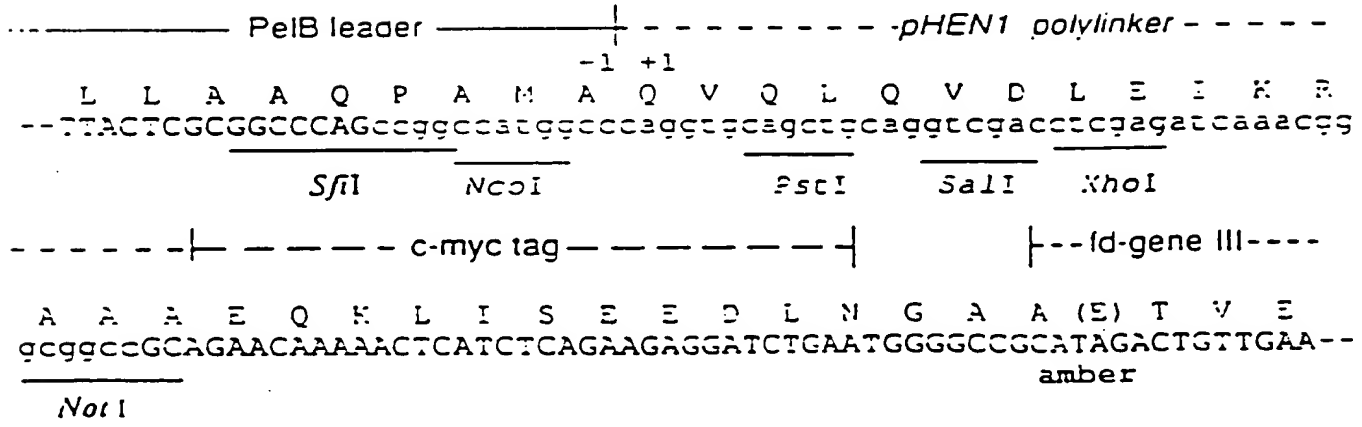


Fig. 27.

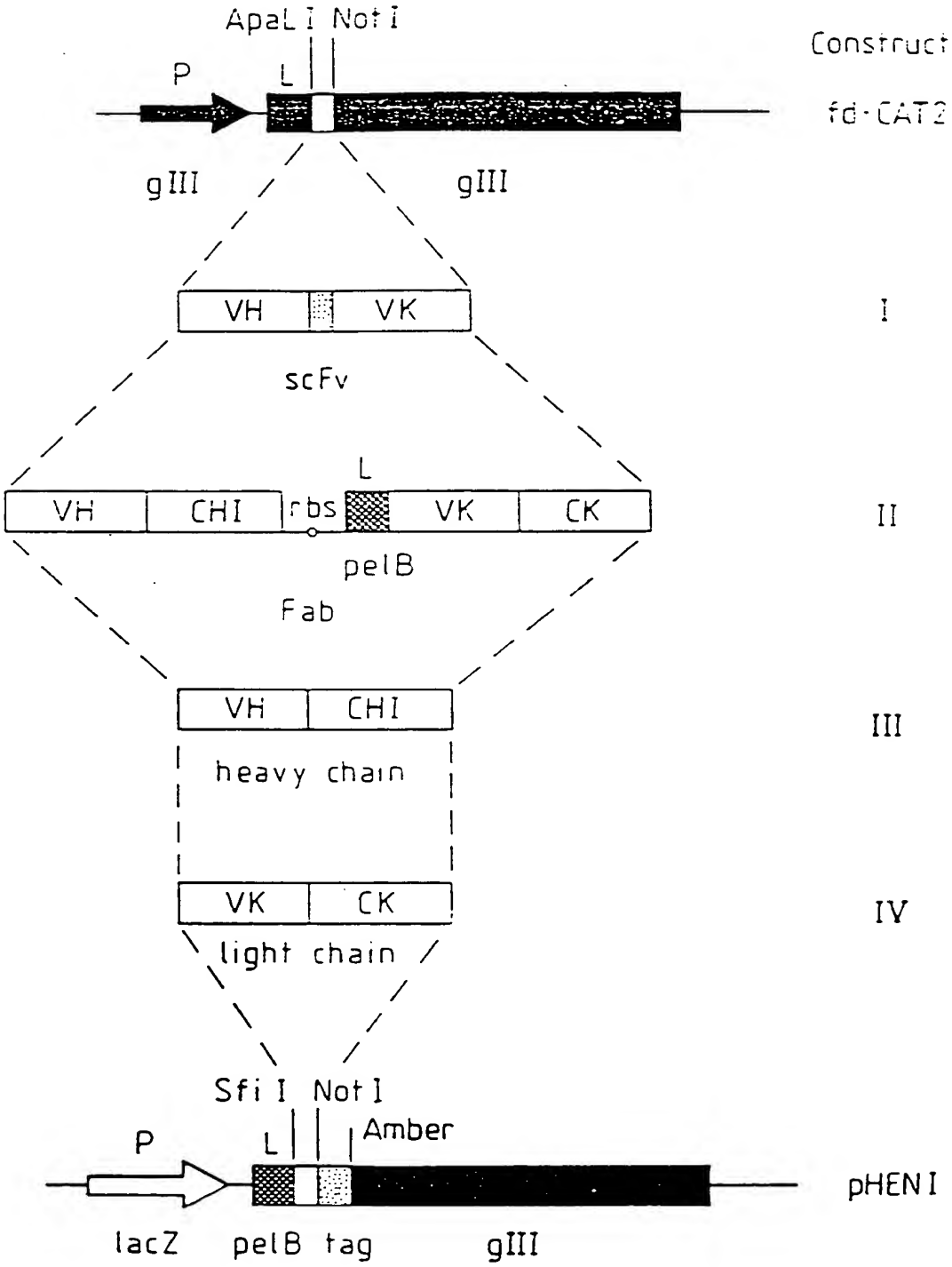


Fig. 28.

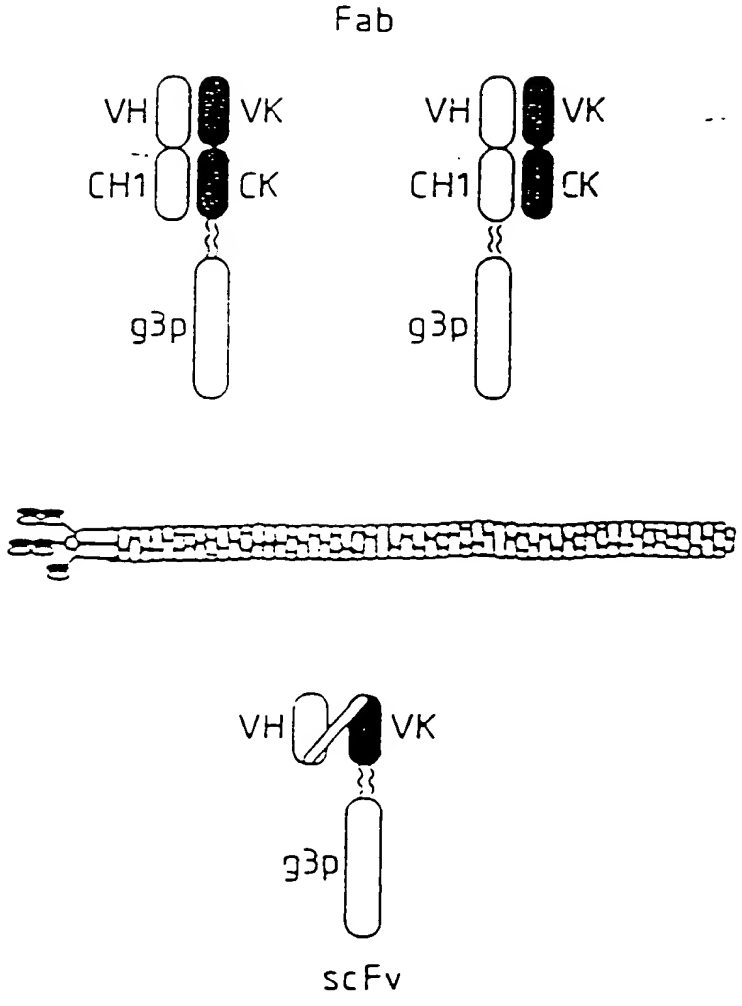


Fig.29.

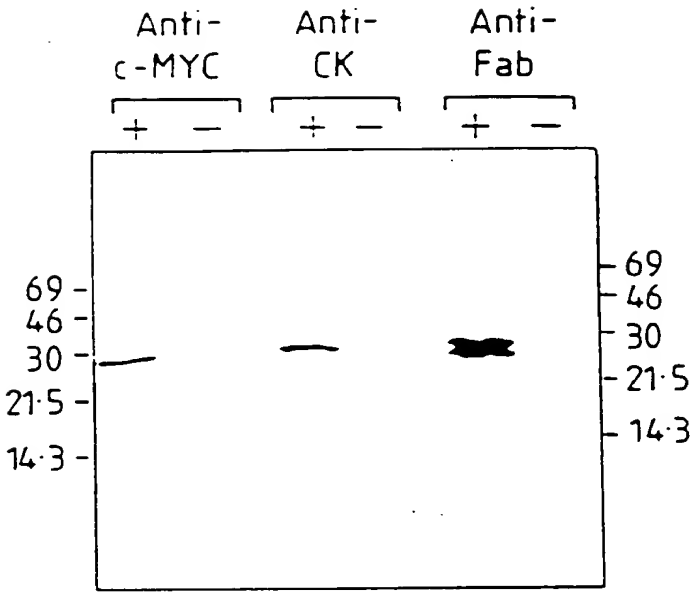


Fig.30.

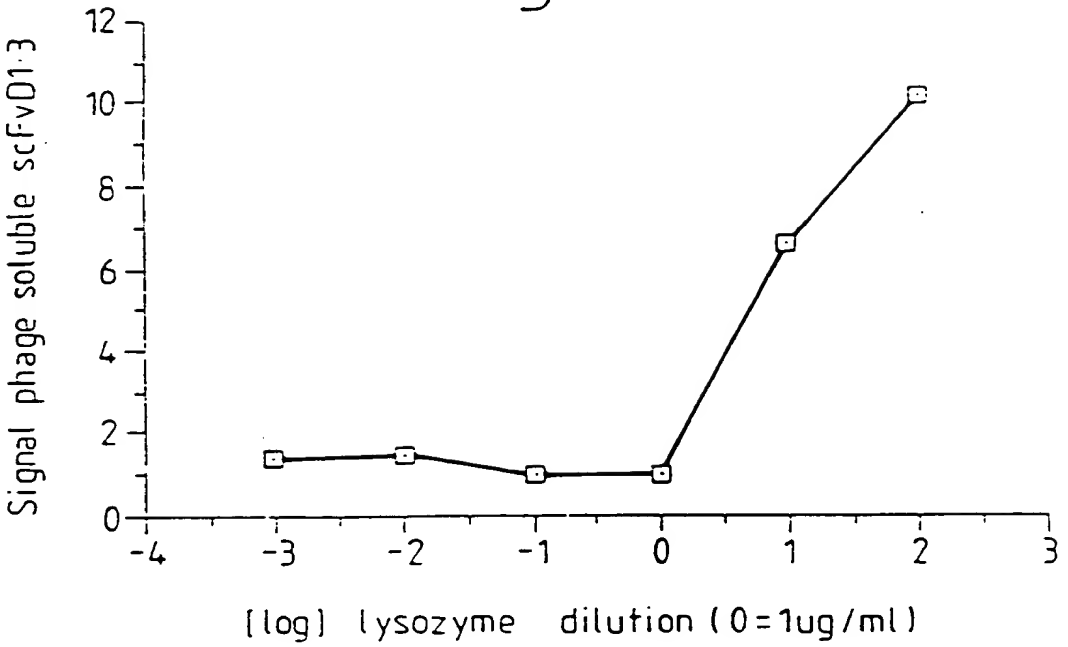

Signal phage soluble scFvD1·3
12
10
8
6
4
2
0
-4
-3
-2
-1
0
1
2
3
[log] lysozyme dilution (0=1ug/ml)

Fig.31.

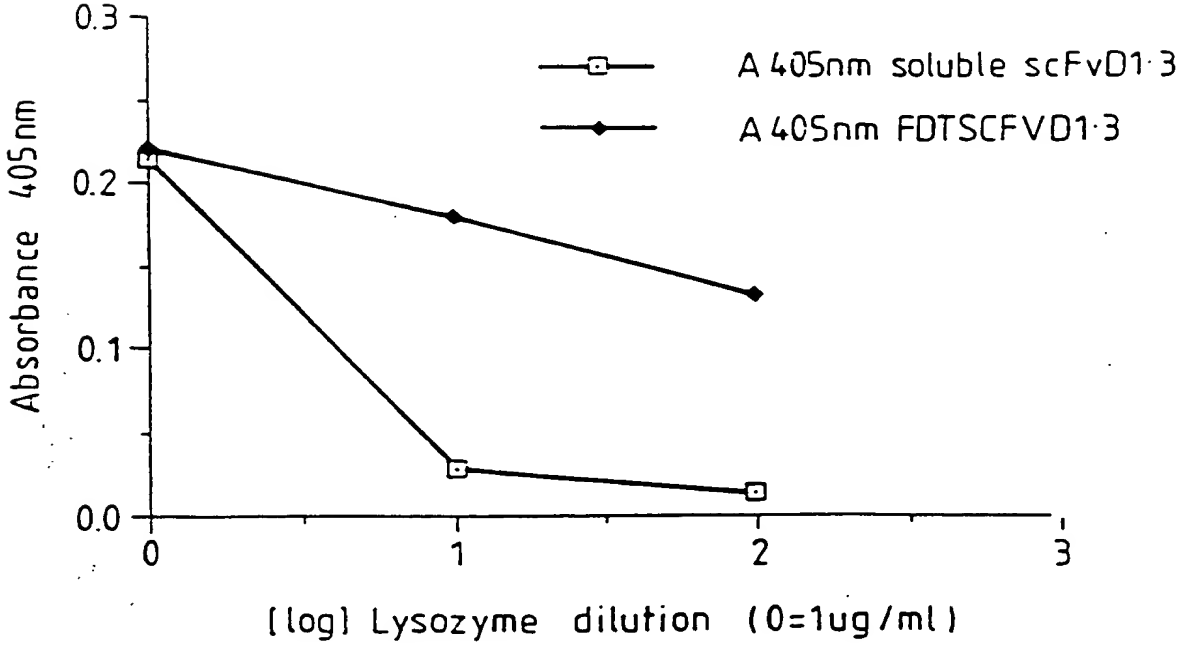

A 405nm soluble scFvD1·3
A 405nm FDTSCFVD1·3
Absorbance 405nm
0.3
0.2
0.1
0.0
0
1
2
3
[log] Lysozyme dilution (0=1ug/ml)

Fig. 32.

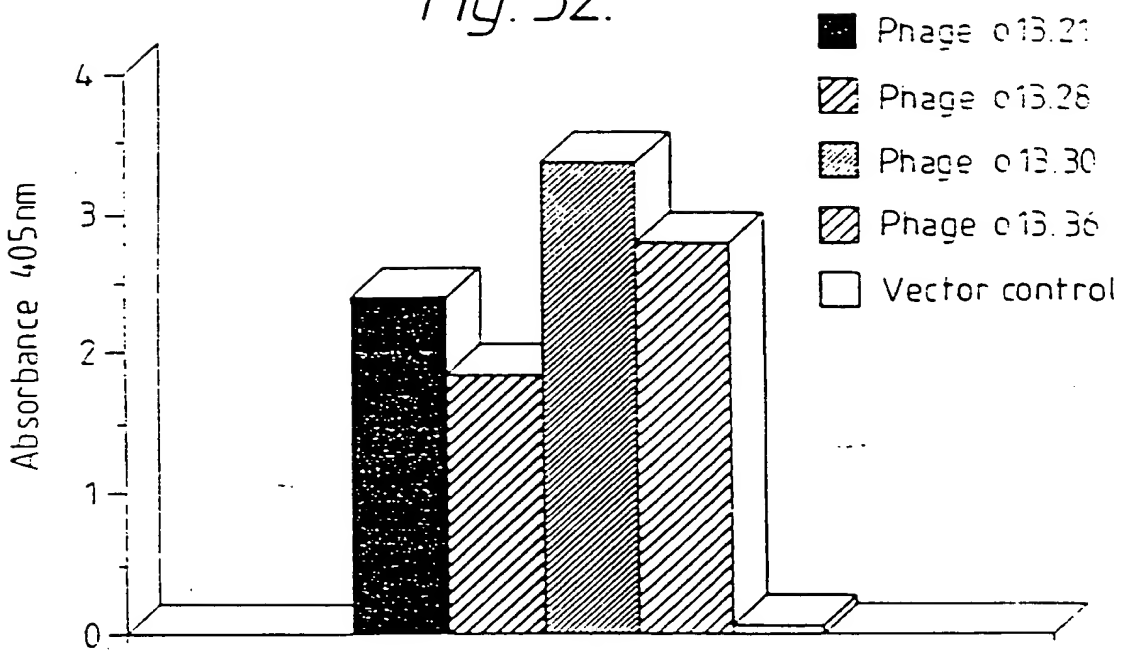

Absorbance 405nm
0
1
2
3
4
Phage o13.21
Phage o13.28
Phage o13.30
Phage o13.36
Vector control

Fig. 33.

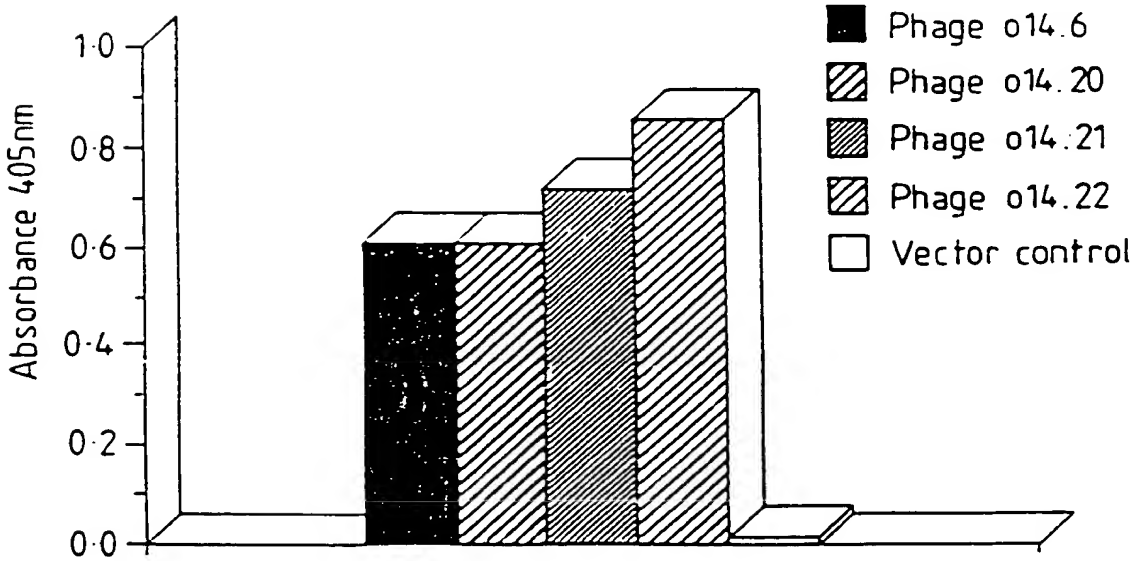

Absorbance 405nm
0·0
0·2
0·4
0·6
0·8
1·0
Phage o14.6
Phage o14.20
Phage o14.21
Phage o14.22
Vector control

Fig. 34.

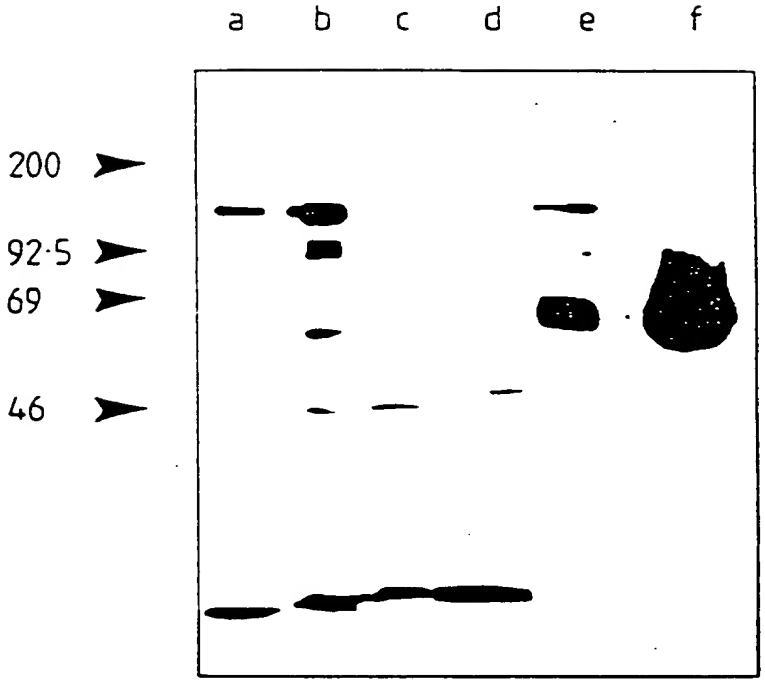


Fig. 35.

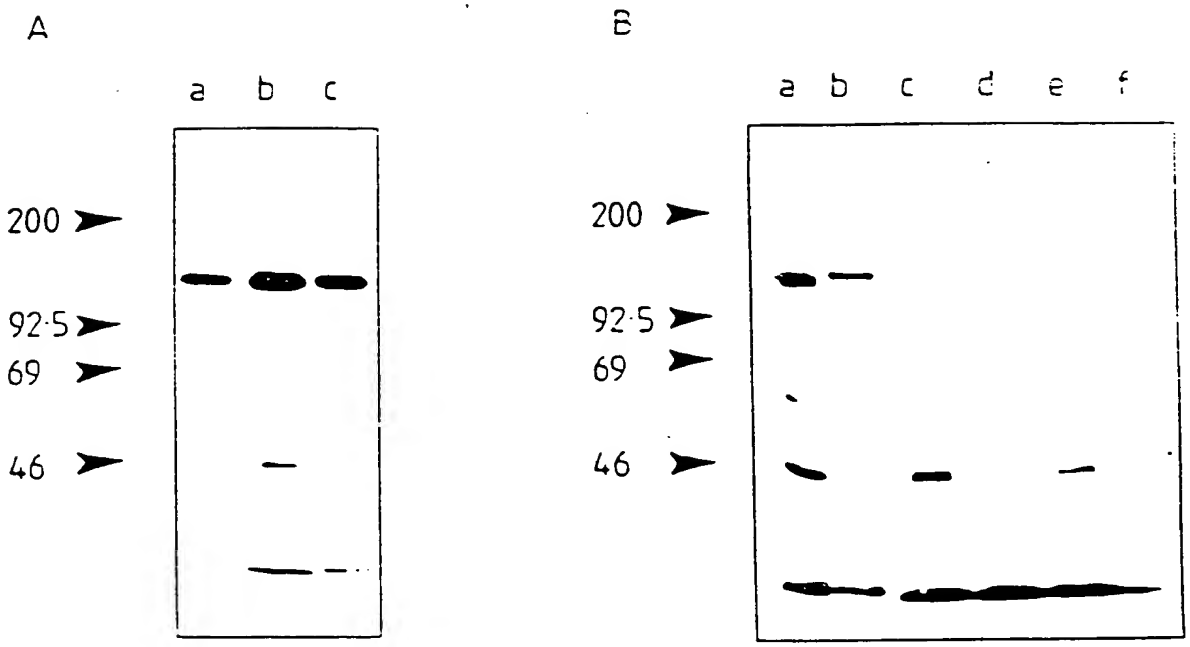
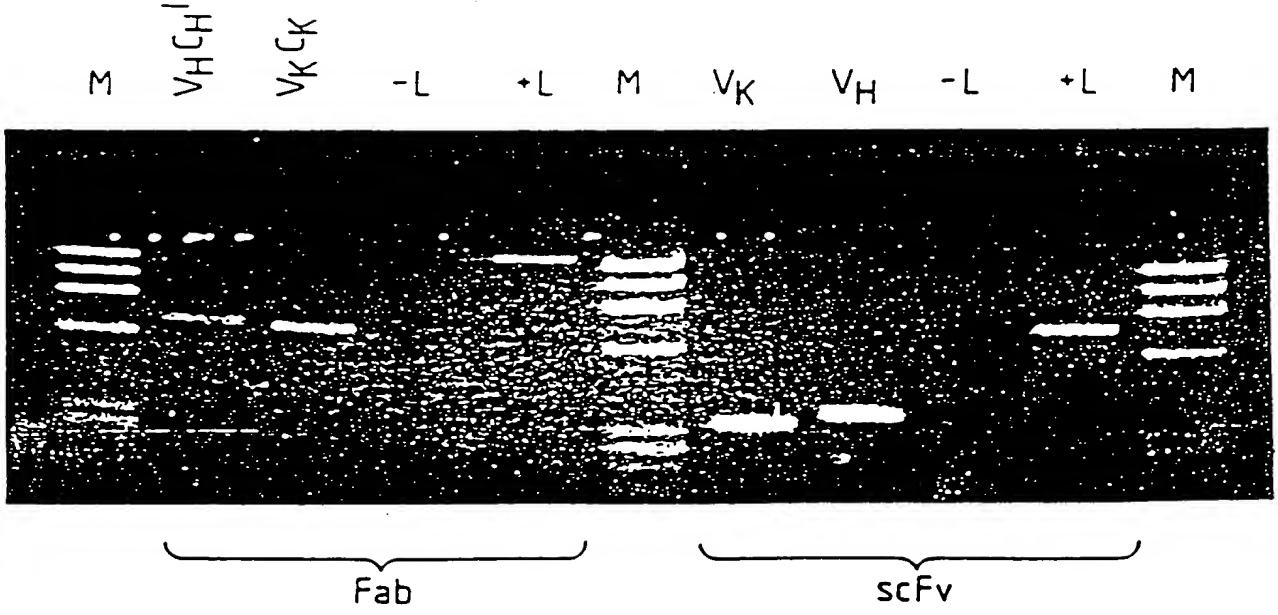


Fig. 36.



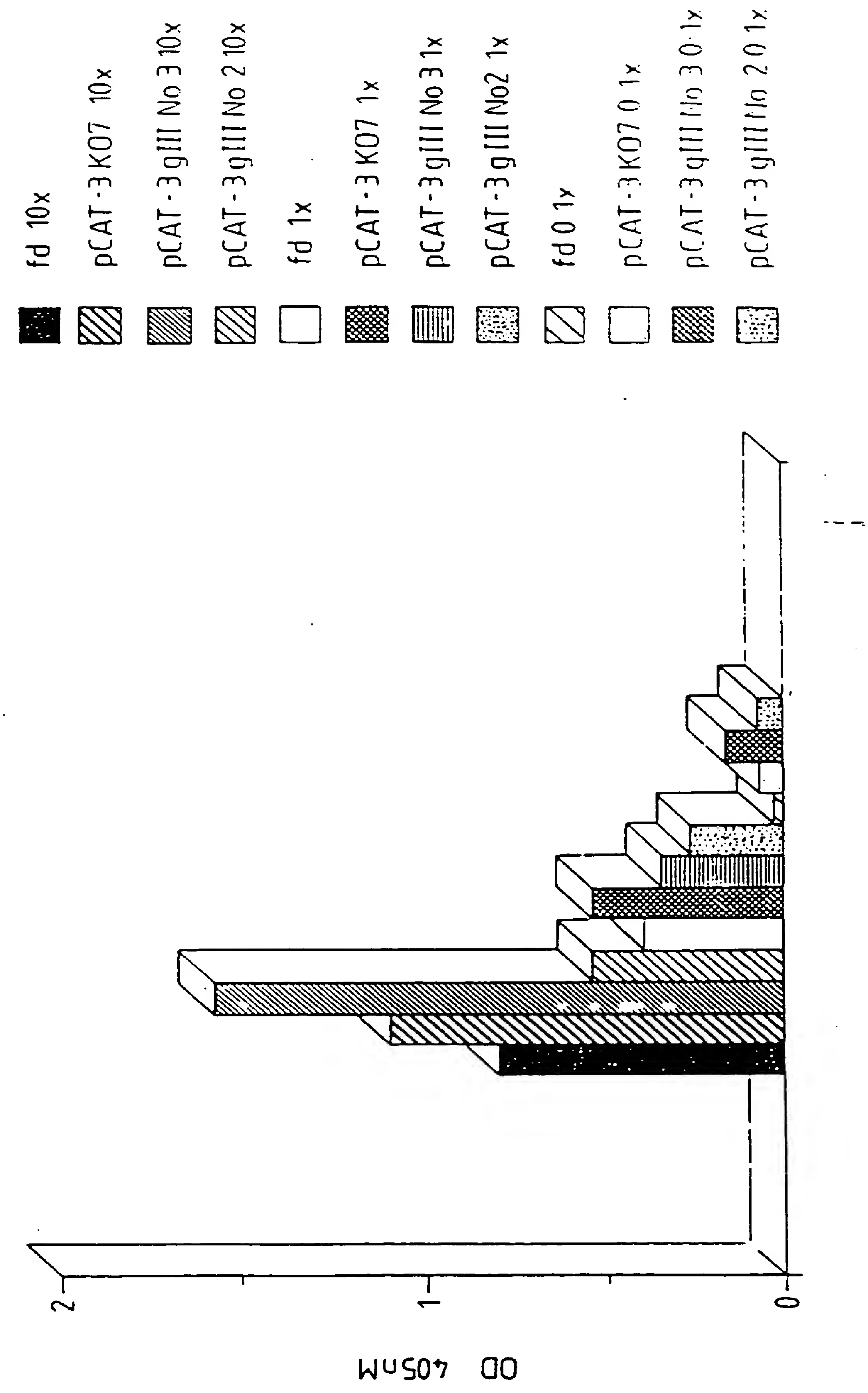

fd 10x
pCAT-3 KO7 10x
pCAT-3 gIII No 3 10x
pCAT-3 gIII No 2 10x
fd 1x
pCAT-3 KO7 1x
pCAT-3 gIII No 3 1x
pCAT-3 gIII No2 1x
fd 0 1x
pCAT-3 KO7 0 1x
pCAT-3 gIII No 3 0·1x
pCAT-3 gIII No 2 0 1x
2
1
0
OD 405nM

Fig.37.

Fig.38.

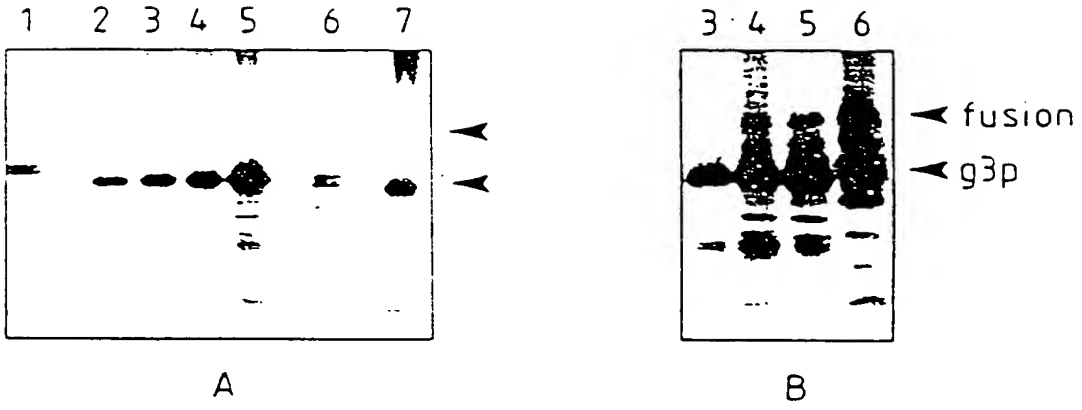


Fig. 39.

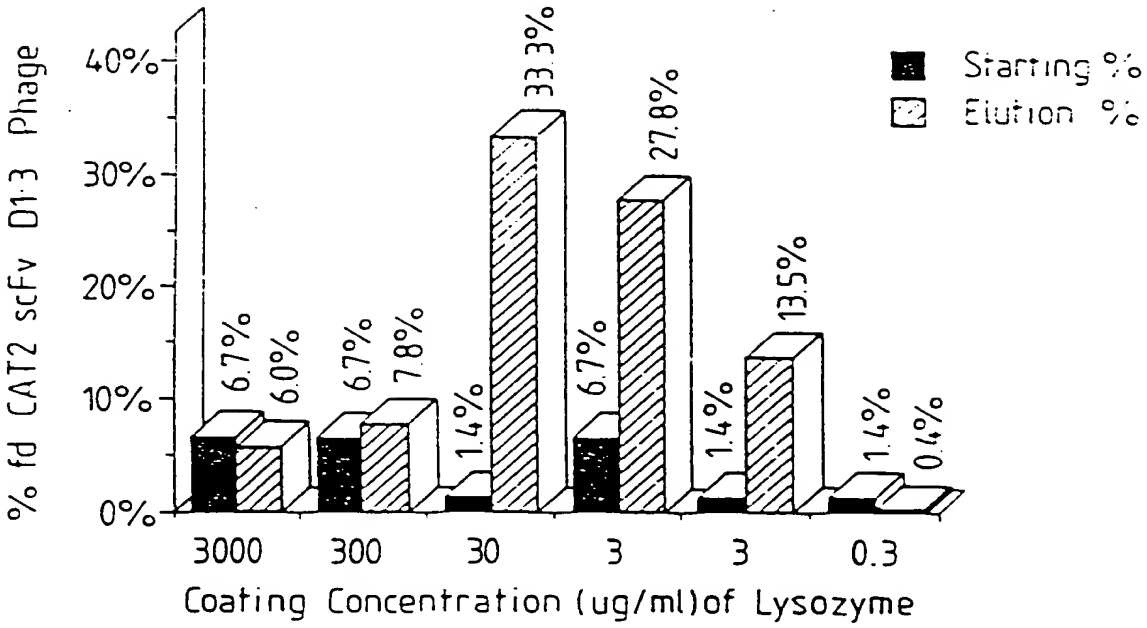

% fd CAT2 scFv D1.3 Phage
40%
30%
20%
10%
0%
6.7%
6.0%
6.7%
7.8%
1.4%
33.3%
6.7%
27.8%
1.4%
13.5%
1.4%
0.4%
3000
300
30
3
3
0.3
Coating Concentration (ug/ml) of Lysozyme
Starting %
Elution %

Fig. 40.

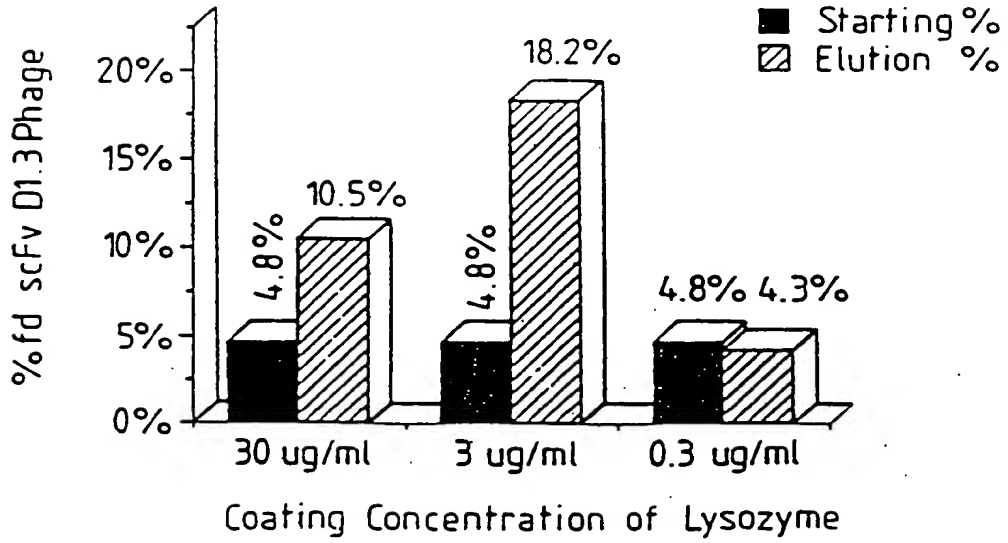

%fd scFv D1.3 Phage
20%
15%
10%
5%
0%
4.8%
10.5%
4.8%
18.2%
4.8%
4.3%
30 ug/ml
3 ug/ml
0.3 ug/ml
Coating Concentration of Lysozyme
Starting %
Elution %

Fig. 41.

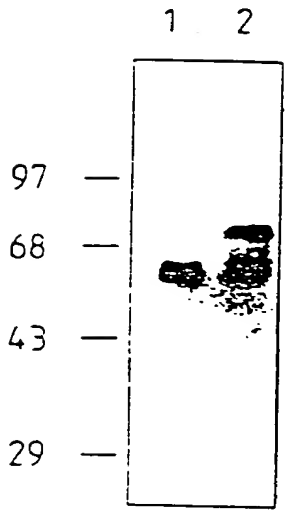


Fig. 42.

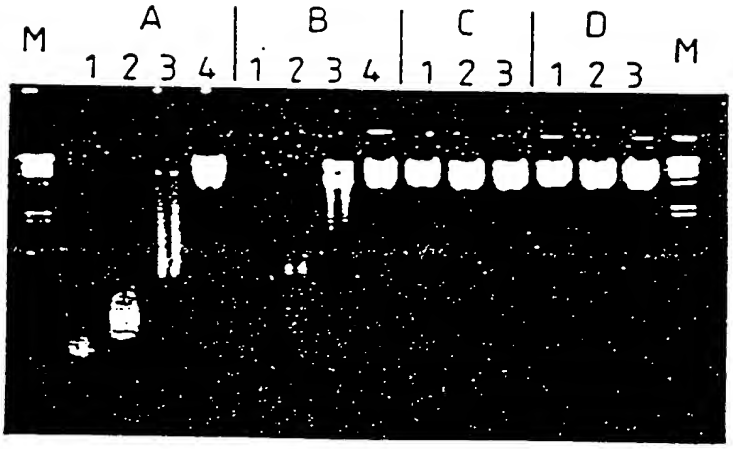


Fig. 43.

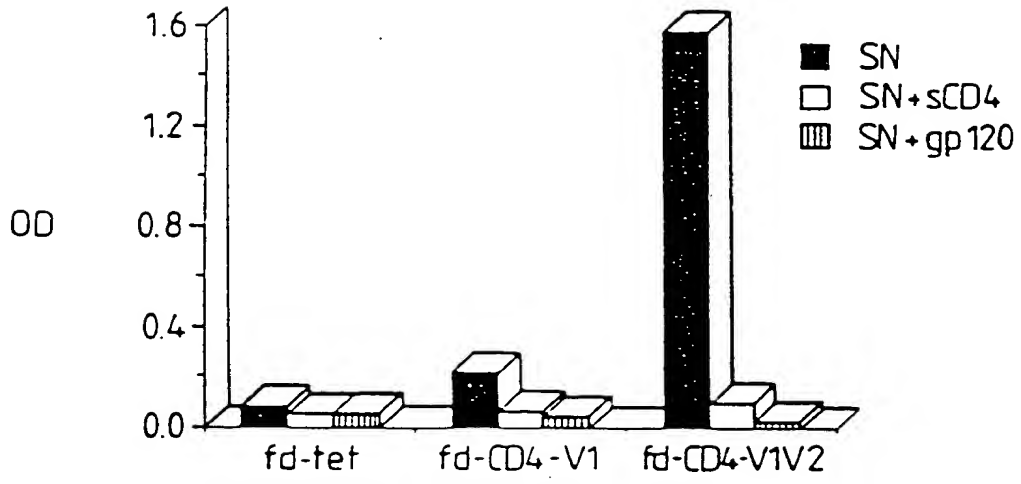


Fig. 44 (i)

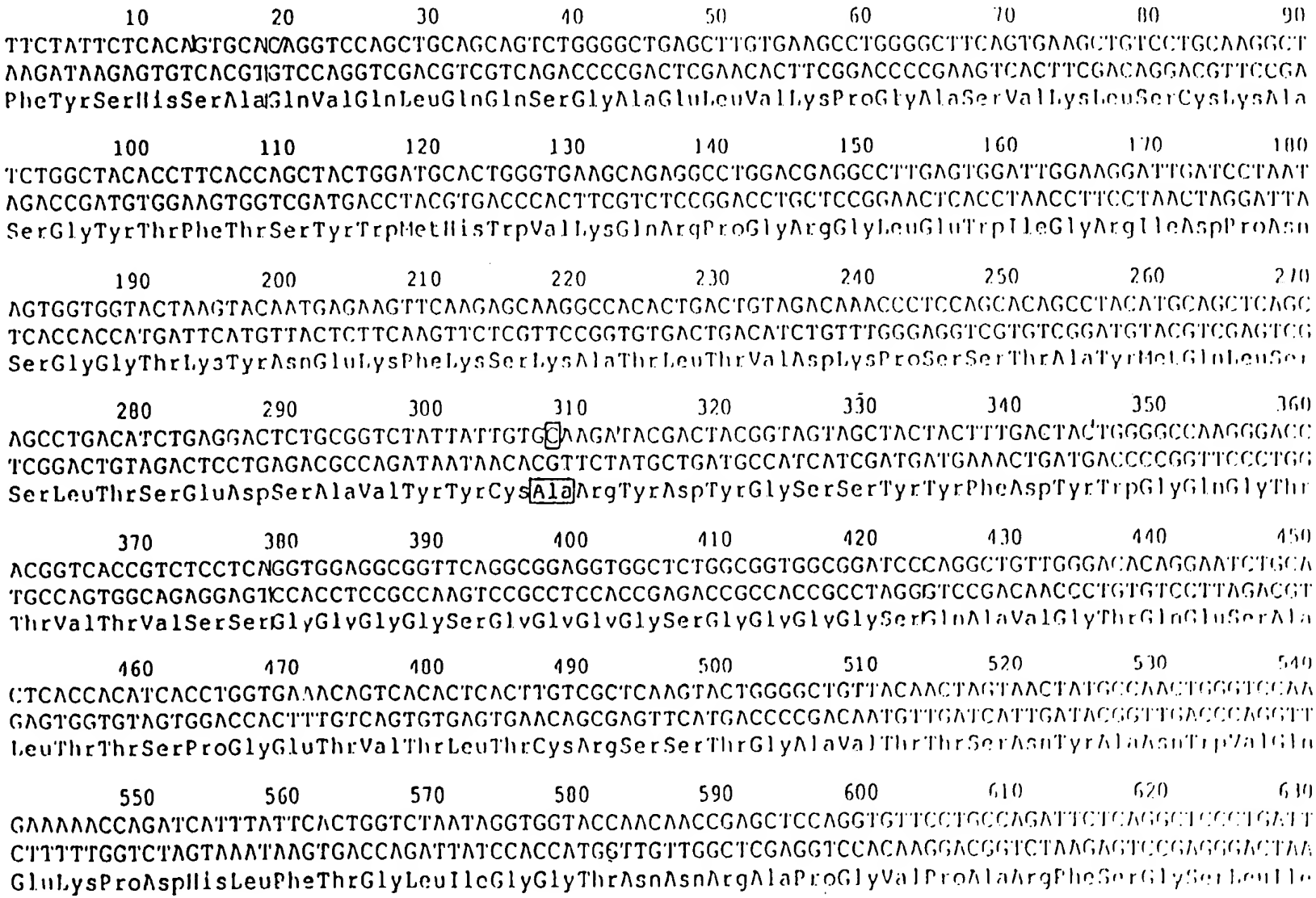


Fig. 44 (ii)

```
           640       650       660       670       680       690       700  C  G 710       720
GGAGACAAGGCTGCCCTCACCATCACAGGGGCACAGACTGAGGATGAGGCAATATATTTCTGTGCTCTATGGTACAGCAACCATTGGGTG
CCTCTGTTCCGACGGGAGTGGTAGTGTCCCCGTGTCTGACTCCTACTCCGTTATATAAAGACACGAGATACCATGTCGTTGGTAACCCAC
GlyAspLysAlaAlaLeuThrIleThrGlyAlaGlnThrGluAspGluAlaIleTyrPheCysAlaLeuTrpTyrSerAsnHisTrpVal

       730       740       750       760       770
TTCGGTGGAGGAACCAAACTGACTGTCCTCGAGATCAAACGGGCGGCCGC
AAGCCACCTCCTTGGTTTGACTGACAGGAGCTCTAGTTTGCCCGCCGGCG
PheGlyGlyGlyThrLysLeuThrValLeuGluIleLysArgAlaAla
```

Fig. 45.

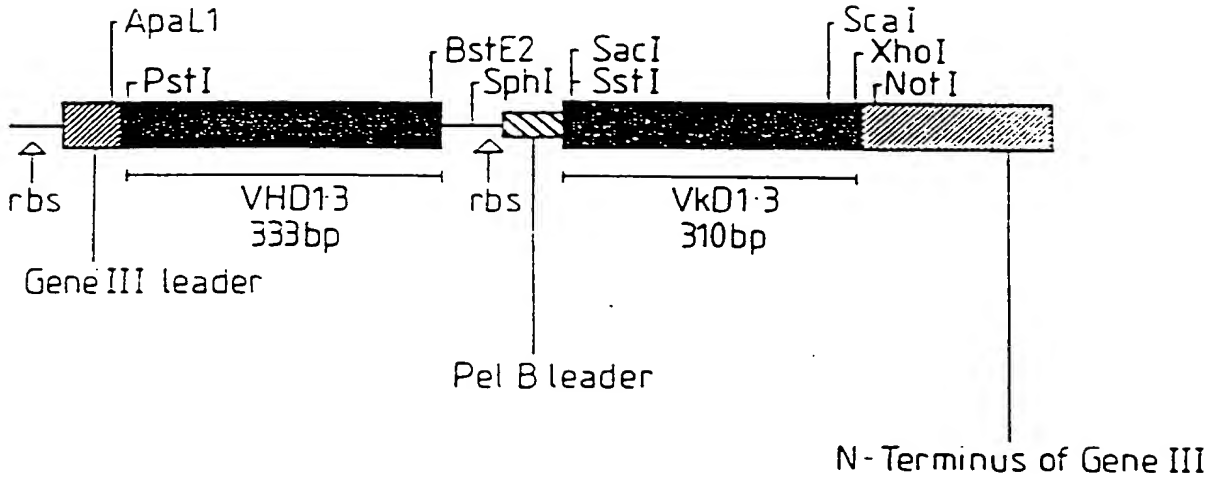

ApaL1
PstI
BstE2
SphI
SacI
SstI
ScaI
XhoI
NotI
rbs
VHD1·3
333bp
rbs
VkD1·3
310bp
Gene III leader
Pel B leader
N-Terminus of Gene III

Fig. 46.

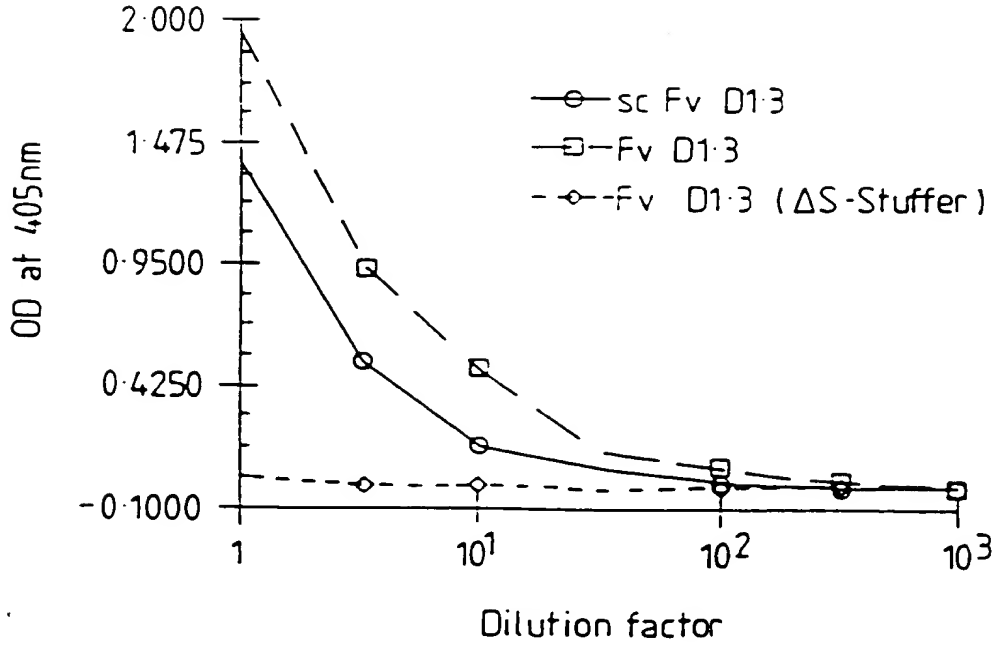

2·000
1·475
0·9500
0·4250
-0·1000
OD at 405nm
sc Fv D1·3
Fv D1·3
Fv D1·3 (ΔS-Stuffer)
1
10¹
10²
10³
Dilution factor

Fig. 47.

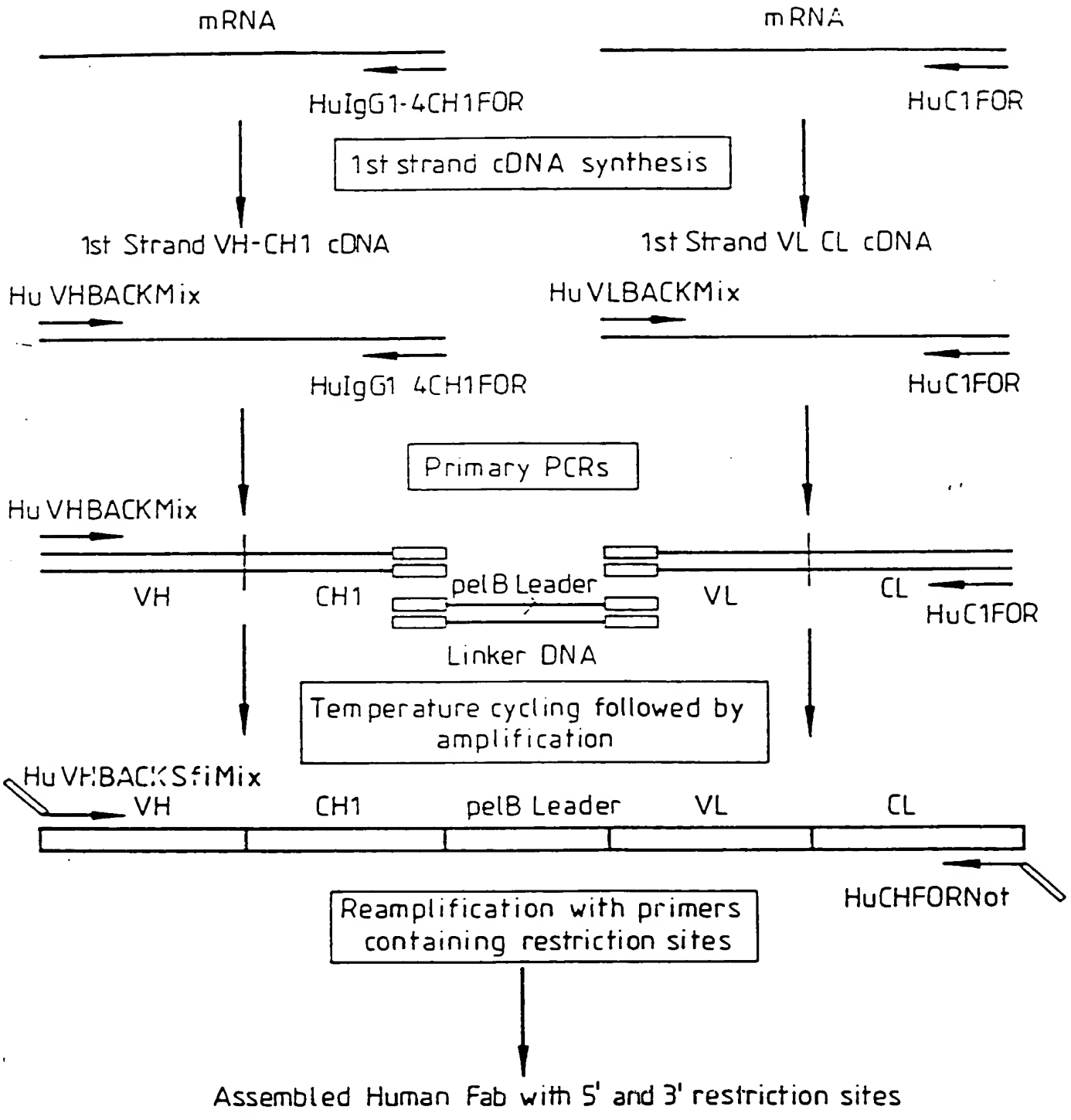
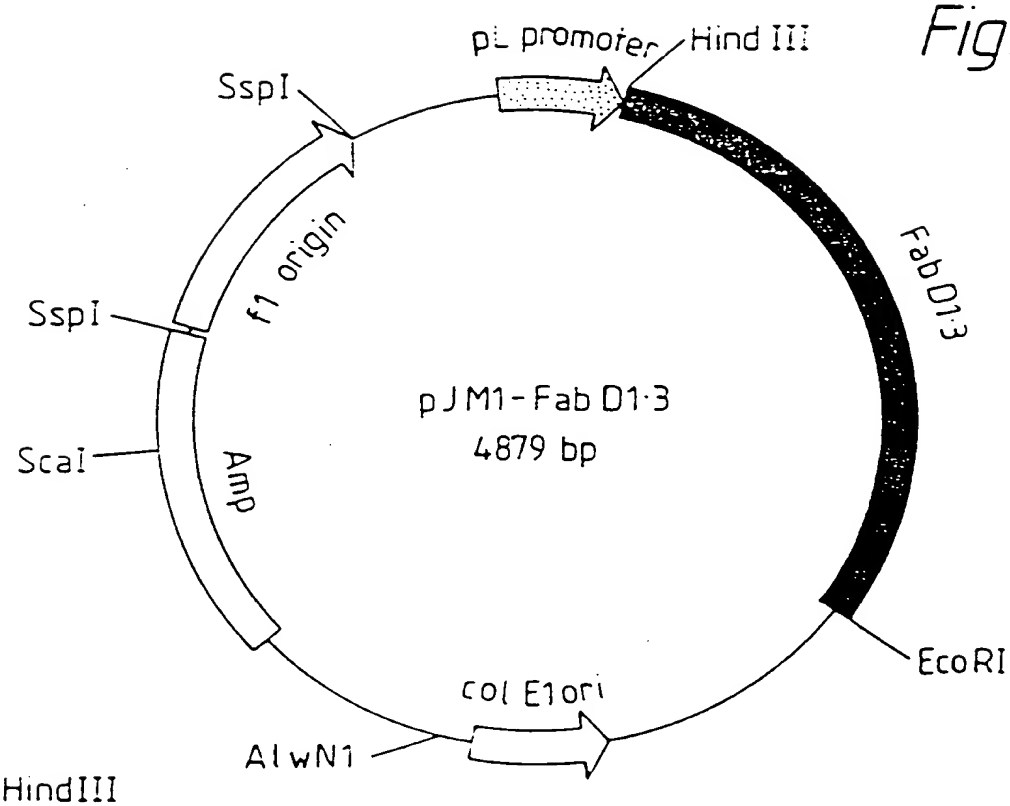
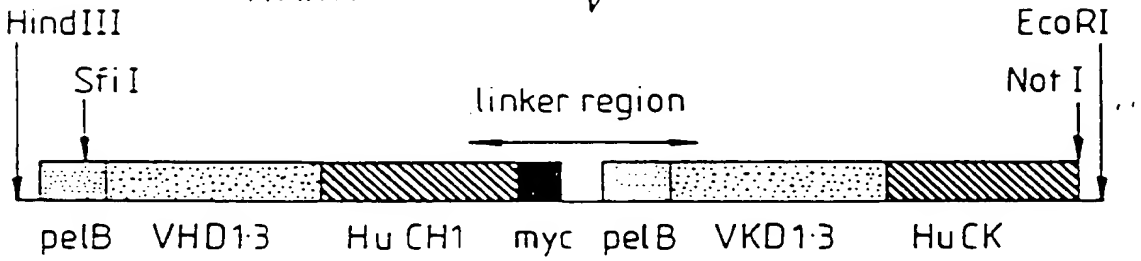


Fig. 48.

A.



B.



C. Sequence of linker region

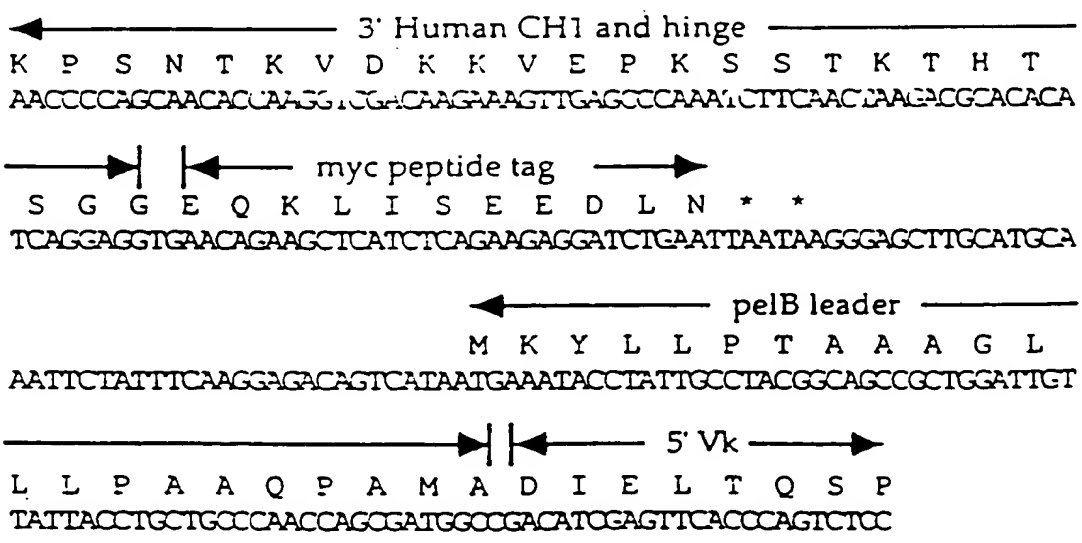


Fig. 49.

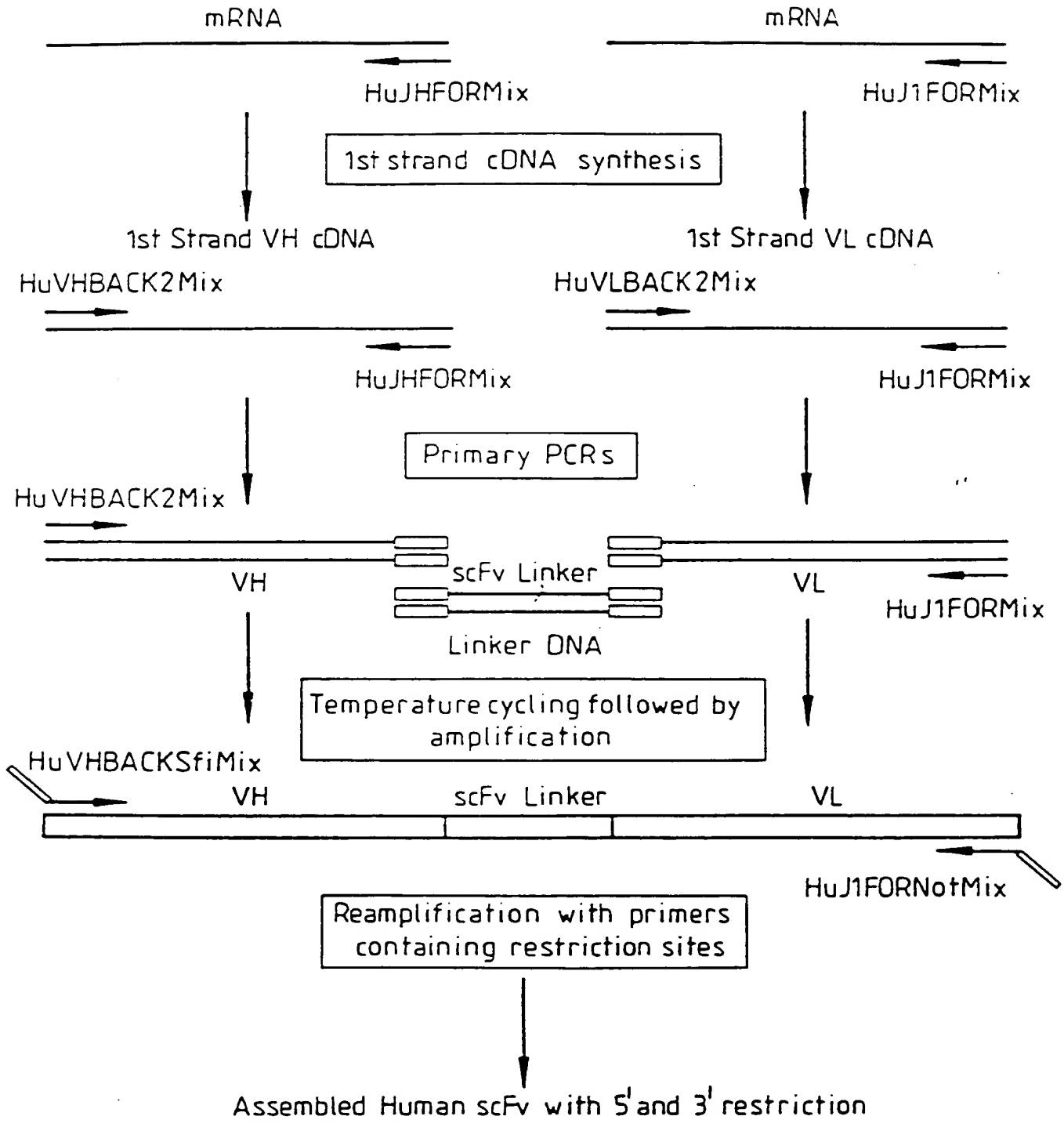
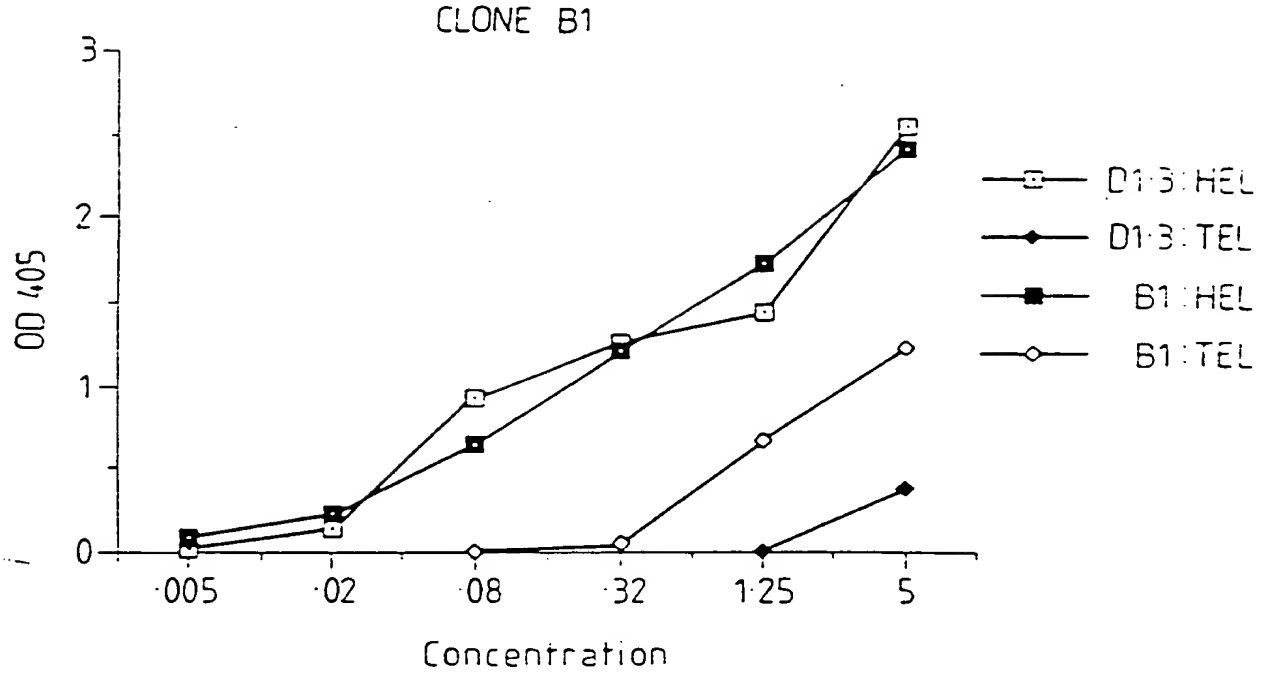


Fig. 50.



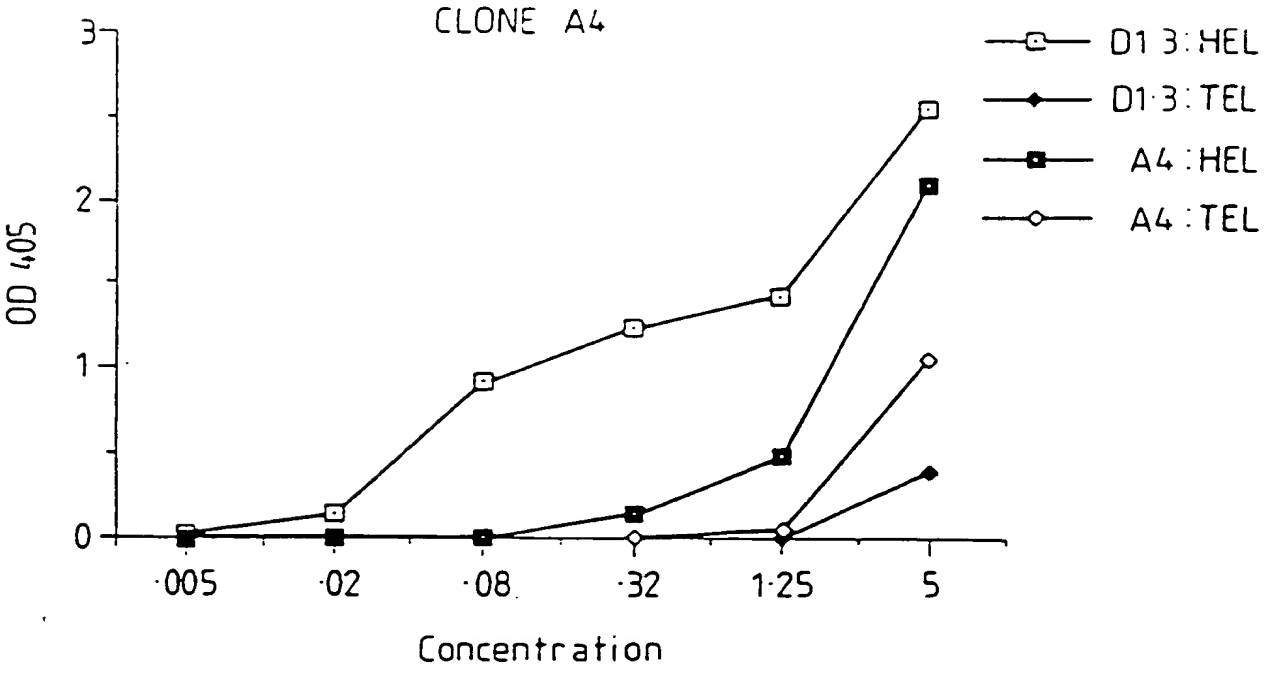


Fig.51.

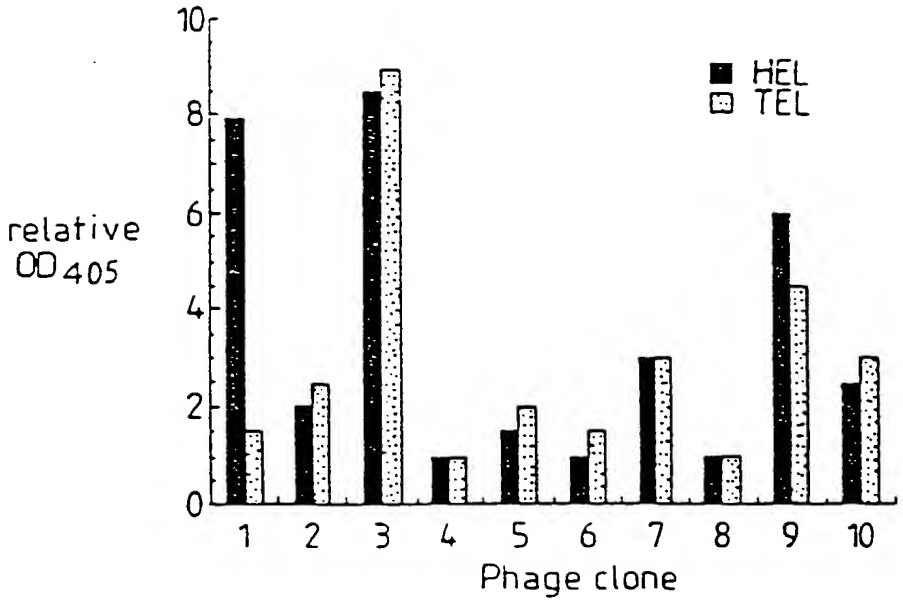


Fig.53.

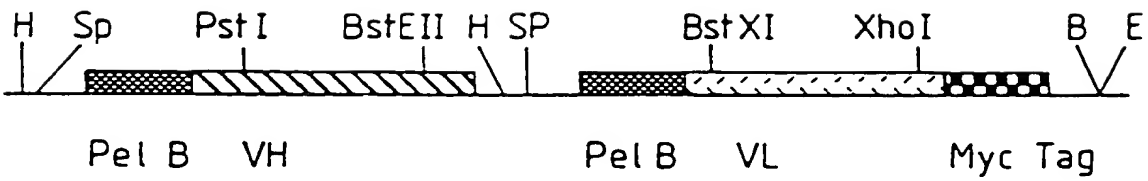


Fig. 52.

	CDR 1	CDR 2
D1.3	DIQMTQSPASLSASVGETVTITCRASGNIHNYLA	WYQQKQGKSPQLLVYYTTTLAD
M1F	DIELTQSPSSLSASLGERVSLTCRASQDIGSSLN	WLQQEPDGTIKRLIYATSSLDS
M21	DIELTQSPAIMAASPGEKVTITCSVSSSISSSNLHWYQQKSETSPKPWIYGTSNLAS	

	CDR 3
D1.3	GVPSRFSGSGSGTQYSLKINSLQPEDFGSYYCQHFWSTPRTFGGGTKLEIKR
M1F	GVPKRFSGSRSGSDYSLTISSLESEDFVDYYCLQYASSPWTFGGGTKLELKR
M21	GVPVRFSGSGSGTSYSLTISSMEAEDAATYYCQQWSSYPLTFGAGTKLEIKR